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**ENVIRONMENTAL QUALITY RESEARCH —  
FATE OF TOXIC JET FUEL COMPONENTS  
IN AQUATIC SYSTEMS**

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## TECHNICAL REVIEW AND APPROVAL

AFAMRL-TR-81-101

The experiments reported herein were conducted according to the "Guide for the Care and Use of Laboratory Animals, "Institute of Laboratory Animal Resources, National Research Council.

This report has been reviewed by the Office of Public Affairs (PA) and is releasable to the National Technical Information Service (NTIS). At NTIS, it will be available to the general public, including foreign nations.

This technical report has been reviewed and is approved for publication.

FOR THE COMMANDER



ROGER C. IMMAN, Colonel, USAF, BSC  
Chief, Toxic Hazards Division



## SUMMARY AND CONCLUSIONS

Preliminary range finding bioassay tests on the water soluble extract (WSF) from the jet fuel JP-4 have been conducted with Daphnia magna, Artemia salina, and the dried eggs of Artemia salina.

Test results with the live organisms tentatively indicated similar sensitivity levels for both species ( $\approx 15-25\%$  diluted WSF).

Both organisms appear to be appreciably more sensitive to the JP-4 WSF than the test fish species, Golden Shiner.

Artemia egg hatchability appears to be dose responsive to JP-4 WSF. However, even control sample hatchabilities have been low in the preliminary tests, reducing the sensitivity of the tests.

A gas chromatographic procedure has been developed for analyzing both the neat JP-4 fuel and its water soluble extract.

Comparative gas chromatograms for several neat JP-4 samples (both petroleum and shale-based) have demonstrated: a) their similarities in terms of components, and b) their differences in terms of relative amounts of individual components.

A procedure for preparing true aqueous solutions (WSF) of JP-4 hydrocarbons has been developed. Preparative factors investigated included fuel/H<sub>2</sub>O ratios and mixing times.

Hydrocarbon composition of the WSF of JP-4, both petroleum and shale-derived, appears to be relatively simple with the major components (benzene, toluene, and xylenes) comprising 70-90 % of the total.

Factors affecting the use of WSF in bioassay tests have been investigated.

A substantial proportion (60-70 %) of WSF hydrocarbons are lost by surface adsorption when stored in glass containers. Pretreatment was shown to eliminate most of this loss.

Biodegradation during WSF storage for limited periods (up to 3 days) appears to be minimal and probably largely limited to the (minor) alkane components.

Preliminary work has been completed dealing with the concept of the mathematical model to be used. The major effort has been directed toward the model format. At this point, for the sake of simplicity, the model will have four compartments: 1) the water soluble aqueous phase, 2) the emulsion aqueous phase, 3) the surface film, and 4) suspended solids.

A literature search reveals the existence of a number of other oil spill models which contain information appropriate to jet fuel spill conditions.

## PREFACE

The research reported herein was conducted at the Sanitary Engineering and Environmental Health Research Laboratory, University of California under the terms of contract F 33615-80-C-0512, Work Unit 63020417, with the U. S. Air Force. The contract monitor was Major J. M. Livingston, Air Force Aerospace Medical Research Laboratory, Wright-Patterson AFB, Ohio. Professor Robert C. Cooper was the Principal Investigator. Dr. Leon Hunter was the project manager. Ms. P. C. Ulrichs and Mr. Robert Okazaki were responsible for the bioassay development.

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## INTRODUCTION

The primary goal of the Berkeley-SEEHRL contract is to investigate the fate of toxic fuel components in the aquatic environment. Unfortunately during the current year the hiatus resulting from the drastic changes in personnel which occurred over a period of several months has resulted in the uneven development of the different but related areas of investigation. As an example in the temporary absence of any manpower to develop and conduct procedures for preparing and analyzing the water soluble extracts of the jet fuel JP-4, tentative and very preliminary attempts were made to develop suitably scaled bioassays using Daphnia magna and Artemia salina. These tests had to be conducted without knowledge of the true levels of the hydrocarbons being evaluated, a point which involved even greater uncertainties than was then realized (due to hydrocarbon losses during storage). Nevertheless, these experiments do provide useful guidance with regard to the design of the upcoming more fully integrated series of tests which will yield reliable data on the toxicity of JP-4 water soluble extract and its individual components.

The development of suitable procedures for preparing and analyzing JP-4 hydrocarbons in true water solution started somewhat late in the contract year as a result of the personnel turnover but is now capable of providing accurate data on hydrocarbon levels, individual and combined, for a variety of tests (bioassays, surface adsorption, evaporative loss, etc.). A number of interesting facts have emerged including a) the variable composition of different JP-4 samples and b) the adsorptive losses of water soluble hydrocarbons.

Late in the year a start was made on evaluating Artemia salina egg hatchability as a bioassay test of hydrocarbon toxicity. The advantages of test simplicity, shelf storability of the dry eggs, etc. make this a potentially attractive system. Unfortunately preliminary tests, described in this report, have been hampered by low hatchability levels (even in controls).

A literature survey of Artemia publications, conducted in conjunction with the hatchability bioassay, provided information of sufficient general interest to justify its inclusion in the present report.

## BIOASSAY DEVELOPMENT

The need for changes in the bioassay procedures (flagfish, aufwuchs, etc.) developed in the previous contract period was foreseen early in the report year. The primary reason was the natural assumption that testing methods would have to be scaled down substantially to accommodate the much smaller test samples to be expected from the fractionated jet fuels and their corresponding water soluble extracts. Accordingly, attention was focussed on smaller aquatic organisms, namely Daphnia magna and Artemia salina, which could be handled in smaller volumes and for which there already existed a substantial volume of published data regarding their suitability as bioassay test organisms.

This section reports the results of early preliminary tests with these organisms. Also reported here for convenience (although started much later) are the Artemia-hatchability tests, also of a preliminary nature.

Biodegradation of the test aqueous hydrocarbons is a potential threat to obtaining meaningful data from the bioassays. Several biodegradation experiments with water soluble extracts in (test) organism-free systems are reported, using dissolved oxygen (DO) as a measure of biodegradation.

## DAPHNIA

A brief preliminary range finding study was conducted. Bioassay guidelines were taken from Standard Methods (1975) and ASTM (1980). Daphnia magna, the "giant water flea" is an aquatic metazoan that appeared suitable for our purpose because of its small size, parthenogenic reproduction, tolerance of low DO, easy availability, and the fact that it lends itself to a variety of studies, e.g. developmental, pathways, enzymatic, and the like.

## Materials

A 100% water soluble fraction (WSF) of JP-4 (batch identified by date, 2-13-79) was formulated by shaking 1 part fuel with 15 parts of aerated Richmond Field Station well water for 2 minutes. This mixture was allowed to separate and stand for 18 hours or longer. The aqueous phase was removed for testing.

As noted in the Introduction, this procedure was used before a suitable analytical technique was developed to monitor water soluble hydrocarbon levels. Based on a subsequent single analysis (see page 28) of this type of WSF taken near the end of the series, it is probable that hydrocarbon levels were substantially below "saturation" (i.e., equilibration) point.

## Methods

Continuous Flow Bioassay. Initial efforts were directed toward a continuous flow system for two reasons. One was based on the report by Orr (1980) that Daphnia exposed to continuous flow dynamics produced greater number of offspring. The second was based on the assumption we would have need of such a system to continually replace volatile material.

After considerable development effort on this system the work was discontinued because a) the animals failed to reproduce, b) maintenance of adequate food concentration was difficult (probably the reason for reproductive failure), and c) the procedure was too cumbersome to be used conveniently.

Static Bioassay. It was decided to develop a clone of D. magna under static growth conditions. The stock was started from specimens received from Ward's Natural Science Establishment, Inc., Rochester, N.Y. To collect young animals for experiments, 50 containers were set up in the following fashion. One D. magna < 24 h old was placed in a container with 100 ml of autoclaved well water. The parents were transferred to a new container after each brood was produced. Neonates from the second and subsequent broods were collected for bioassays. The first brood was not used because these were often smaller (Green, 1954), weighed less (Goulden, 1980), and might respond differently to toxicants. Feeding was based on the formula of LaMotte: 6 mg/l final concentration of Fleishmann's live cake yeast plus 1.5 mg/l final concentration of Kordon's Artemia food — fed Monday through Thursday and three times this amount on Friday. Under these conditions the

stock remained parthenogenic and showed normal growth: 1st brood was released at 10 days of age and the 2nd, 3rd, and 4th broods at 3-day intervals. At 21°C longevity was 4 weeks for 78% (based on 19 individuals), 5 weeks for 17% (based on 41 individuals).

For the toxicity tests the water soluble extract was diluted with aerated Richmond Field Station well water. The test chambers were filled to the top (leaving no air space) and capped. They were not opened until the end of the experiment or until all test organisms died, at which time DO measurements were taken. Nominal WSF of fuel concentrations of 0, 5, 8, 12, 20, 31, 50, and 65% in well water were tested. Ten animals were exposed to 300 ml of solution in stoppered BOD bottles at a temperature of 23°C. The pH of the solutions was 8.7 and the DO concentration at zero time was 8.3 mg/l. The bioassay was continued for 144 h. The animals did not survive under control conditions for much more than 72 h, indicating a maximum 48-h test endpoint.

### Results

The results of the preliminary *D. magna* bioassay study are presented in Table 1 and the corresponding tentative no-effect level (NOEL) values are summarized in Table 2 and compared with similar data for Golden Shiners exposed to JP-4 in a previous study (Klein et al., 1976). These preliminary data appear to indicate that *D. magna* is appreciably more sensitive to JP-4 than is the Golden Shiner.

TABLE 1  
PRELIMINARY *Daphnia* BIOASSAY STUDY  
Exposed to JP-4 Batch Date 2-13-79

Nominal Conc. % WSF	Survival					Dissolved Oxygen, mg/l				
	Time in Hours					Time in Hours				
	0	24	48	72	144	0	24	48	72	144
0	10	10	10	10	0	8.3 <sub>b</sub>	-	-	-	7.7
0	10	10	10	10	0	-	-	-	-	7.7
5	10	10	10	10	5	-	-	-	-	3.0
8	10	10	10	0	0	-	-	-	-	2.3
12	10	10	10	0	0	-	-	-	6.3	-
20	10	0	0	0	0	-	-	7.6	-	-
31	10	0	0	0	0	-	7.9	-	-	-
50	10	0	0	0	0	-	7.9	-	-	-
65	10	0	0	0	0	-	7.9	-	-	-
100 <sup>a</sup>						7.9	-	-	-	1.1

<sup>a</sup> No test animals.

<sup>b</sup> Dashes indicate no DO measurements available.

TABLE 2

COMPARISON OF JP-4 NO-EFFECT LEVEL (NOEL) BETWEEN  
D. magna AND THE GOLDEN SHINER

	NOEL <sup>a</sup>		
	24 h	48 h	72 h
<u>D. magna</u>	< 12	< 12	< 5
Golden Shiners <sup>b</sup>	< 32	< 20	< 20

<sup>a</sup> NOEL expressed as % WSF of fuel

<sup>b</sup> Data ex Klein et al., 1976

ARTEMIA (BRINE SHRIMP)

Artemia is a salt water organism with many advantages similar to Daphnia. In addition, the nauplii are easily obtained from the dried eggs or cysts. The latter, readily available from San Francisco Bay Brand, Newark, California, can be shelf-stored for months so that it is unnecessary to maintain a continuous culture stock. (See Appendix A for further information.)

The preliminary bioassay tests were intended to provide guidance in the following areas:

- a) to obtain some idea of the sensitivity of Artemia to the toxic jet fuel components (for comparison with other aquatic organisms, especially Daphnia).
- b) to determine the feasibility of scaling down the test volumes to accommodate limited amounts of jet fuel fractions.
- c) to determine the appropriate time periods required to obtain LC 50 data.

Materials and Methods

The jet fuel water soluble fraction was prepared from JP-4 (Batch 2-13-79) as described in the D. magna section (page 9 ).

The tests were conducted at 3 volume levels: 1.5 ml, 11.5 ml, and 300 ml (standard BOD bottle). The Artemia (3 ml of eggs) were hatched in 3 liters of 2.8% saline water, transferred after 24 h, and aged for a further 24 h before testing. In general, 10 Artemia were used per test. In a few cases in the second test series, a larger number (50) were used to assess possible effects of crowding. The test water was Richmond Field Station well water containing 28 g Leslie rock salt per liter and the appropriate levels of the jet fuel water soluble components. In addition to mortality measurements, the DO levels were intermittently measured in the large-scale (BOD bottle) tests. Two sets of experiments were carried out in which the jet fuel water solubles

were obtained from water-jet fuel mixes that were allowed to separate for 1 and 2 days, respectively.

### Results

1. The data in Tables 3 and 5 yield the approximate no-effect level (NOEL) values shown in Table 7.
2. The NOEL data differ substantially in the two test series, presumably as a result of the variation in time allowed for settling in the preparation of the water soluble extract.
3. Forty-eight hours would appear to be the minimum test period for the determination of LC 50 values (not calculated from present tentative data).
4. Comparisons with NOEL data for Daphnia are inconclusive as regards relative sensitivity to jet fuel. Even using the more "reliable" Test 2 series the 24-h data suggest lower sensitivity and the 48-h data higher sensitivity of the Artemia — perhaps a result not too unexpected from this type of ranging experiment.
5. The test results with larger numbers of animals were too erratic to allow definite conclusions although the increased mortality (even in the control) in the small-volume experiments was suggestive of overcrowding.

### BIODEGRADATION

One factor which must be controlled and/or evaluated in any bioassay of jet fuel toxicity is the potential biodegradability and hence removal of the fraction of soluble extract being tested. Conflicting evidence for and against biodegradation based on DO levels in salt and fresh water has shown up in the bioassay experiments with Artemia and Daphnia. For example, in the 100% (i.e., undiluted) WSF samples, which contained no test animals, a substantially greater DO drop with time was observed in the fresh water system (Table 1) compared with the salt water (Tables 4 and 6). It was decided to investigate this further in a brief series of tests using the same WSF at three levels (0, 45, and 90%) in salt and fresh water. As before, DO values were used as a measure of biodegradation.

### Results

The only evidence for a change in oxygen concentration in the salt water system is a slight dip in DO at the intermediate jet fuel extract concentration after 120 h (Table 8). On the other hand, in fresh water the oxygen loss is substantially greater but still occurs at the 45% fuel extract concentration. Thus, these results confirm the previous evidence for little or at least much less hydrocarbon loss in salt water. In fresh water, biodegradation takes place but is substantially inhibited at higher WSF concentrations, presumably because of the increased toxicity toward the degrading organisms. The resulting "dip" in DO levels (and presumably in degrading water soluble fractions) indicates the need for care in establishing appropriate conditions for our bioassay tests (although the O<sub>2</sub> losses are much less at the shorter test period of 48 h.

TABLE 3

Artemia BIOASSAY - FIRST TEST SERIES

(Aqueous Extract) Nominal Conc. %	No. of <u>Artemia</u> in Test	Survival Number								
		1.5 ml			11.5 ml			300 ml		
		Test Volume			Test Volume			Test Volume		
		Time, h			Time, h			Time, h		
		24	48	120	24	48	120	24	48	120
0	10	10	10	0	10	10	0	10	10	0
0	10	10	10	0	10	10	0	10	10	0
5	10	10	0	0	10	10	0	10	10	0
8	50	0	0	0	50	50	0	50	50	0
12	10	10	10	0	10	10	0	10	10	0
20	35	0	0	0	35	35	0	35	35	0
31	10	10	10	0	10	10	0	10	5	0
50	39	39	0	0	39	0	0	39	0	0
65	10	10	0	0	10	0	0	10	0	0

TABLE 4

OXYGEN LEVELS IN Artemia BIOASSAY - FIRST TEST SERIES

Nominal Concentration % Aqueous Phase of JP-4	Dissolved Oxygen, mg/l ; Time in Hours			pH Initial
	0	48	120	
0	7.3	-	6.8	8.35
0	- <sup>b</sup>	-	6.7	-
5	-	-	5.1	-
8	-	-	3.9	-
12	-	-	3.8	-
20	-	-	1.8	-
31	-	-	2.5	-
50	-	5.6	0.5	-
65	-	6.2	5.4	-
100 <sup>a</sup>	7.0	6.4	6.8	8.05

<sup>a</sup> This sample contained no test animals

<sup>b</sup> No measurements made on samples marked "-"



TABLE 5

Artemia BIOASSAY - SECOND TEST SERIESArtemia Survival Number for Test Chambers  
of the Following Capacity:

Nominal Conc. % Aqueous Phase of JP-4	No. of <u>Artemia</u> in Each Volume	1.5 ml			16.0 ml			300 ml		
		Time in Hours			Time in Hours			Time in Hours		
		24	48	72	24	48	72	24	48	72
0	11	11	10	6	12	6	1	12	10	1
8	10	10	9	5	10	5	3	10	7	2
14	10	10	6	3	10	6	3	11	2	0
24	10	9	5	3	10	7	5	10	4	0
42	10	8	4	3	5	4	2	0	0	0
75	10	0	0	0	1	0	0	0	0	0
0	50	2	0	0	50	43	29	50	25	1
24	50	49	0	0	49	42	2	50	0	0
42	50	50	0	0	50	24	4	12	0	0

TABLE 6

OXYGEN LEVELS IN Artemia BIOASSAY - SECOND TEST SERIES

Nominal Concentration % Aqueous Phase of JP-4	Dissolved Oxygen, mg/l				pH	
	Time in Hours				Time in Hours	
	0	24	48	120	0	120
0	7.5	-	-	6.6	8.10	8.10
8	- <sup>b</sup>	-	-	5.8	-	8.15
14	-	-	-	5.1	-	8.15
24	-	-	-	3.0	-	8.00
42	-	6.9	-	2.1	-	7.90
75	-	6.9	-	5.1	-	8.05
0	-	-	-	6.6	-	8.15
24	-	-	6.6	2.4	-	-
42	-	-	6.4	3.0	-	7.95
100 <sup>a</sup>	7.3	-	6.8	6.8	8.02	8.05

<sup>a</sup> This sample contained no test animals<sup>b</sup> No measurements made on samples marked "-"

TABLE 7  
NO-EFFECT LEVEL VALUES FOR Artemia

Test Volume ml	NOEL <sup>a</sup>			
	Test 1		Test 2	
	24 h	48 h	24 h	48 h
1.5	b	b	14	0
11.5 (or 16.0)	100	31	24	0
300	100	20	24	0

<sup>a</sup> Expressed as % WSF of fuel

<sup>b</sup> Data too erratic to estimate NOEL

<sup>c</sup> Even the 0% control showed a 54% kill.

It should be noted that the above evidence for biodegradation does not appear to be supported by the tests described in a later section of this report (page 31) in which hydrocarbon levels were monitored directly in WSF stored in the presence and absence of the antibacterial  $\text{HgCl}_2$ . However, the difference may be more apparent than real since biodegradation may only involve the alkane hydrocarbons which are a very minor part of the WSF and whose reduction therefore will cause only a small, statistically insignificant change in the measured total hydrocarbon levels.

#### Use of Antibiotics to Prevent Biodegradation

The possible use of antibiotics to inhibit biodegradative effects on the WSF of JP-4 in fresh water was examined. The test measured the changes in dissolved oxygen over a three-day period of seven nominal WSF concentrations (0, 12, 20, 32, 50, 79, and 100%) with and without antibiotics. Penicillin (100 units per ml, final concentration) along with streptomycin (0.1 mg/ml, final concentration) were used.

The data are presented in Table 9. The antibiotics inhibit degradation for the 72-h period (no significant DO change). In the test series without antibiotics the DO pattern was similar to the previous experiment: the  $\text{O}_2$  losses were much less at the 48-h time period, but at 72 h there was a progressive DO drop up to a fuel concentration of 100%.

Since the antibiotics seem to be effective in controlling biodegradation, their effect on the test organism, Daphnia, was studied. Conditions were similar to the DO test.

Unfortunately, as the data in Table 10 show, the antibiotics were toxic to Daphnia. Over a 24-h period less than half survived.

TABLE 8

COMPARISON OF DO CHANGES IN FRESH AND SALT  
WATER SOLUTIONS OF JP-4 WATER SOLUBLES

	Nominal Concentration % Aqueous Phase of JP-4	Dissolved Oxygen, mg/l		
		Time in Hours		
		<u>0</u>	<u>48</u>	<u>120</u>
SALT WATER	0	7.70	7.50	7.40
	0	7.70	7.50	7.40
	45	7.75	7.60	7.10
	45	7.80	7.60	7.10
	90	7.75	7.60	7.70
	90	7.80	7.70	7.40
FRESH WATER	0	9.00	9.10	9.10
	0	9.00	9.10	8.90
	45	9.00	8.85	0.65
	45	9.00	8.75	3.50
	90	9.00	8.90	7.90
	90	9.00	8.90	8.00
TEMPERATURE, °C		22.3	20.5	20.5

TABLE 9

COMPARISON OF DO CHANGES IN WSF DILUTIONS  
WITH AND WITHOUT ANTIBIOTICS

	Nominal Concentration % WSF of JP-4	Dissolved Oxygen, mg/l			
		Time in Hours			
		<u>0</u>	<u>24</u>	<u>48</u>	<u>72</u>
ANTIBIOTICS	0	8.7	8.8	8.9	8.9
	12	8.5	8.6	8.7	8.6
	20	8.6	8.6	8.6	8.6
	32	8.6	8.7	8.7	8.7
	50	8.5	8.6	8.7	8.7
	79	8.5	8.6	8.6	8.6
	100	8.7	8.6	8.3	8.3
NO ANTIBIOTICS	0	8.8	8.8	8.8	8.8
	12	8.5	8.3	7.8	7.2
	20	8.6	8.4	7.7	6.9
	32	8.7	8.5	8.0	6.8
	50	8.7	8.8	8.5	6.1
	79	8.7	8.6	8.4	5.3
	100	8.6	8.6	8.3	8.0

TABLE 10  
COMPARISON OF Daphnia BIOASSAYS WITH  
AND WITHOUT ANTIBIOTICS

	Nominal Concentration % WSF of JP-4	Survival Time in Hours		
		0	24	48
ANTIBIOTICS	0	10	4	0
	12	10	0	0
	20	10	0	0
	32	10	0	0
	50	10	0	0
	79	10	0	0
NO ANTIBIOTICS	0	10	10	10
	12	10	9	9
	20	10	1	0
	32	10	0	0
	50	10	0	0
	79	10	0	0

#### Artemia HATCHABILITY BIOASSAY

The hatchability of cysts (dry eggs) of brine shrimp *A. salina* has been used successfully in testing environmental contaminants (Kuwabara et al., 1980). These preliminary studies were conducted to determine the feasibility of using this hatchability methodology as a simple and rapid test in assessing jet fuel toxicity.

#### Materials and Methods - Test Series I

The cysts were obtained from the University of California, Bodega Marine Laboratory. The cysts originated from South San Francisco Bay. After 1 mg of cysts ( $\approx$  300 eggs) were placed in each of ten glass vials, 10 ml distilled water were added to each vial. The vials were capped and then stored at 4°C for 10 h. Conte et al. (1977) reported the elimination of hatching asynchrony by prehydration of cysts at low temperatures (3°C) for 4 to 10 h in distilled water. After this prehydration period, the cysts were collected by Millipore filtration (0.45  $\mu$ m). The ten filters were allowed to air dry.

A 100% WSF of JP-4 (Batch GEC-1A-792033) was prepared by the procedure described on page 27. After a 24-h settling period, the WSF was used in making the following volumetric percent concentrations: 25%, 50%, 75%, and 100%. These concentrations were prepared in duplicates, as well as the controls (0%). The test container was a 130-ml glass bottle fitted with a ground glass wide-mouth hollowed mushroom stopper. Fifty-ml samples of WSF were analyzed chromatographically as described on page 23.

After sampling for hydrocarbon analysis, the filters containing the cysts were carefully placed in each bottle and carefully mixed for three sec to disperse the cysts and then the filter immediately removed. The bottles were

incubated at 25°C for 48 h. After this time period, hatchability counts were performed. The following two categories were selected for the count procedure: 1) early prenauplius with hatching membrane and its connection to the cyst (E<sub>1</sub> stage) and late prenauplius with hatching membrane and separation from cyst (E<sub>2</sub> stage), and 2) free-swimming nauplius. (See Appendix A for description of Artemia life history.) Successful hatchability was considered the free swimming nauplius stage. A baseline hatchability value (nauplii count/mg cysts) was determined from the control samples. The nauplii number per mg cysts in each of the test concentrations was then compared with this baseline value to obtain a relative hatchability percentage.

#### Results - Test Series I

The results of the GC analysis of the WSF of JP-4 are presented in Table 11.

TABLE 11  
RESULTS OF GC ANALYSIS WSF OF JP-4 - TEST SERIES I

<u>Nominal % Volumetric Concentration</u>	<u>Measured Concentration ppm Total Hydrocarbon</u>	<u>Actual % Concentration</u>
0	0	0
25	4.6	16
50	13.6	48
75	21.5	76
100	28.3	100

The total hydrocarbon concentration for the 100% WSF was 28.3 ppm. Analyzed concentrations for 75% and 50% dilutions were in excellent agreement to the expected values; however, the concentration for 25% dilution was about 35% lower than expected. These values were initial concentrations and should be assumed as relative since storage experiments (see page 31) demonstrated losses in the 100% WSF after 48 h.

The hatchability results are tabulated in Table 12. Results between the duplicates vary considerably in the control and lowest fuel concentrations (25%). One duplicate from the 25% WSF concentration showed greater nauplii count than the controls. However, nauplii counts decreased with increasing fuel concentrations in the three highest WSF exposures. An interesting observation was that there generally was a greater prenauplii count (E<sub>1</sub> + E<sub>2</sub> stages) in the 25% and 50% WSF than in the controls. The effects of the WSF of JP-4 on cyst hatchability are clearly illustrated in Figure 1. The percent relative hatchability showed > 80% reduction at 50% WSF and 100% reduction at 75% and 100% WSF.

The above results indicated the feasibility of this test in assessing jet fuel toxicity. However, methodologies needed to be worked out to minimize variability between duplicates. The 300 cysts/mg approximation initially

TABLE 12

HATCHABILITY COUNT PER MILLIGRAM CYST AFTER 48-HOUR  
INCUBATION PERIOD AT 25°C

	Pronauplii Stage (E <sub>1</sub> + E <sub>2</sub> )	Nauplii Stage
0% A*	13	28
0% B*	75	73
25% A	35	129
25% B	101	38
50% A	14	6
50% B	101	10
75% A	1	1
75% B	0	0
100% A	0	0
100% B	0	0

\* Duplicate samples

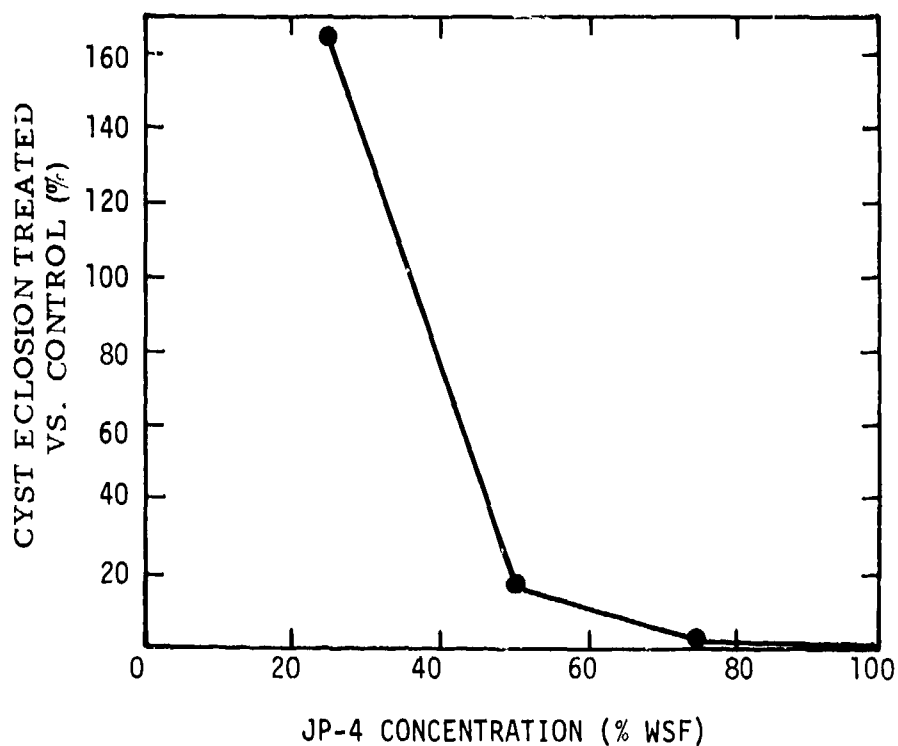


FIGURE 1. HATCHABILITY OF *Artemia* CYSTS EXPOSED  
TO FOUR CONCENTRATIONS OF WSF OF JP-4

contributed to this variability. Two 1-mg cyst samples were later counted and showed wide variations, i.e., 243 and 440 cysts. In addition, numerous cysts were observed to be floating at the air-water interface; these cysts may have been nonviable, as well as empty cyst shells. In the second test series only "settled" cysts in known numbers were used. Furthermore, since WSF hydrocarbon concentrations were now known to decrease by 60-70% during storage (see page 31) steps were taken to study and attempt to minimize hydrocarbon losses.

#### Materials and Methods — Test Series II

As stated previously, to minimize cysts floating at the surface, settled cysts only were used in this experiment. A glass column, 1.2-m long and 3.8-cm diameter fitted at the bottom with a glass stopcock, was filled with distilled water. Approximately 1 g of cysts was placed in the water and settled cysts were collected after about 15 min. Two-hundred cysts were counted and put into 10 ml distilled water in each of the 16 glass vials. After the vials were capped they were stored at 4°C for 5-7 h.

A 100% WSF of JP-4 was prepared as described in the previous section. All glassware used in preparing the various test concentrations and the test containers, 130-ml glass reagent bottles fitted with ground glass stoppers, were presaturated with their respective WSF concentrations for 24 h prior to the commencement of the experiment. This procedure was used in an attempt to minimize fuel loss by adsorption onto the glass surface (see page 33). The following volumetric percent concentrations were prepared in duplicate: 0% (control), 3.2%, 6.5%, 18%, 32%, 42%, and 65%. Fifty-ml diluted samples were analyzed as in Test Series I.

Each 200-cyst batch was collected on a Millipore filter (0.45  $\mu$ m) and then rinsed gently into a bottle with its respective test solution. All bottles were filled to the top and stoppered to prevent formation of air space. The cysts were incubated at 25°C for 66 h. At the end of 66 h, 25 ml were taken from each duplicate concentration and combined to make a 50-ml sample for analysis. Care was taken to exclude cysts or nauplii during sampling. Two blank 65% and 100% WSF (without cysts) were treated in the same manner as described above to determine hydrocarbon loss after 66 h. Hatchability counts were made after 66 h.

#### Results — Test Series II

The WSF concentrations analyzed by gas chromatography are shown in Table 13. The total hydrocarbon for the 100% WSF of JP-4 is 24.3 ppm. At 0 h the measured concentrations were in close agreement to the expected values. After 66 h WSF concentrations were generally 15-21% lower (see page 35) than the initial values (confirming the beneficial effect of pretreatment of the glassware with WSF).

Hatchability results are presented in Table 14. The experiment was originally planned to terminate after 48 h but very low nauplii counts at this hour in the two controls suggested extending the incubation period another 18 h for more hatching to occur. Unfortunately, after 66 h low hatchability (< 5%) was still observed in concentrations 0% to 18% WSF. At

concentrations 32% to 65% no free nauplii were observed. Duplication was better than the previous experiment.

TABLE 13  
RESULTS OF GC ANALYSIS OF WSF OF JP-4 - TEST SERIES II

<u>% Volumetric Concentration</u>	<u>Measured Conc. ppm 0 h</u>	<u>Actual % Concentration</u>
0	0	0
3.2	0.41	1.7
6.5	1.13	4.7
18.0	4.33	17.8
32.0	7.86	32.3
42.0	10.85	44.7
65.0	16.90	69.5
65.0 Blank	16.90	69.5
100.0 Blank	24.30	100.0

TABLE 14  
HATCHABILITY COUNT PER 200 CYSTS AFTER 66-HOUR  
INCUBATION PERIOD AT 25°C

	<u>Prenauplii Stage (E<sub>1</sub> + E<sub>2</sub>)</u>	<u>Nauplii Stage</u>	<u>% Hatchability</u>
0% A (48 h)	39	9	4.5
0% B (48 h)	29	9	4.5
0% A	29	2	1.0
0% B	36	4	2.0
3.2% A	49	3	1.5
3.2% B	37	8	4.0
6.5% A	44	3	1.5
6.5% B	30	3	1.5
18.0% A	21	2	1.0
18.0% B	21	2	1.0
32.0% A	42	0	0
32.0% B	19	0	0
42.0% A	13	0	0
42.0% B	20	0	0
65.0% A	7	0	0
65.0% B	8	0	0



The low hatchability observed in this experiment was unfortunate. The previous Test Series I showed approximately 24% hatching in one control sample and a high of 43% in one of the lowest fuel exposures. Hatching of less than 5% may be attributed possibly to very low oxygen levels since no air space was provided in the test bottles, as well as using salt-free medium. Clegg (1964) showed that cysts consumed more oxygen as salt concentration decreased and he cites Dutrieu (1960) who stated that oxygen is an absolute requirement for emergence. Oxygenated distilled water may help to improve hatchability in future experiments.

In general the results of these two series of tests show promise as a toxicity bioassay method provided hatchability levels in the controls can be raised substantially to allow for a statistically better spread in the number of nauplii in the test dilutions.

## INVESTIGATION OF JP-4

### ANALYTICAL METHODS

Neat fuels (1  $\mu$ l) were analyzed on a Fisher Series 4800 gas chromatograph with dual FID detectors, using dual 18 ft x 1/8 in. columns packed with 10% SE 30 on Chromosorb W (80/100 mesh). Runs were programmed as follows: 60°C for 10 min, 60°C increasing to 200°C at 16°C/min and finally held at 200°C until all the components had emerged (~12 min). Nitrogen carrier gas flowrates of approximately 50-60 ml/min were individually adjusted to balance column bleed. Peak areas were integrated using an Autolab Minigrator.

Previous procedures for monitoring water soluble jet fuel hydrocarbons have involved multiple solvent extraction and concentration steps, prior to GC analysis, which are not only time consuming but also result in the loss of the more volatile jet fuel components. We have now developed a procedure which allows us to dispense with the tedious solvent removal step while at the same time effecting a ten-fold increase in hydrocarbon concentration, ample for satisfactory quantitation by GC.

The procedure is based on the fact that the distribution of hydrocarbons in a water-pentane two-phase system is heavily weighted toward the organic phase. Consequently, it is readily possible to extract, essentially quantitatively, hydrocarbons from a given volume of aqueous solution using much smaller amounts of pentane. For example, three extractions, each with 1-1.5 ml pentane, will recover 95-98% of the hydrocarbons in 50 ml water soluble extract of a jet fuel. The combined pentane extracts adjusted to a standard 5 ml volume have thus effected a ten-fold concentration of the water soluble hydrocarbons. At this level the hydrocarbons can be readily measured by gas chromatography.

The gas chromatographic procedure is essentially as described except that 10- $\mu$ l samples of the pentane extract were used and the electrometer sensitivity was increased approximately 100-fold. Figure 2 is a chromatogram of a typical water soluble extract. The identified peaks (benzene, toluene, xylenes) were located by the addition of authentic samples. Peaks

were quantitated by adding n-pentadecane as internal standard. In the present report, since no correction factors have been used to compensate for different responses relative to the standard, concentrations are given in "C<sub>15</sub> units." This does not appreciably decrease the accuracy of the analysis since the only hydrocarbons affected, the aromatics, have responses approaching 95% of the C<sub>15</sub> response (Szepesy, 1970).

Due to their greater solubility in water, the aromatic hydrocarbons are the primary constituents of the water soluble extracts, although they are a minor proportion of the whole jet fuel. This fact can be used to monitor for the presence of fuel emulsion in the aqueous phase since fuel droplets will have a composition close to the whole fuel and hence will give a chromatogram with a greatly increased contribution by the alkane components. In practice we monitor the ratio of peak "A" (a small alkane peak, see Figure 2, emerging just prior to the benzene peak) to the benzene peak. In an emulsion-free system peak "A" is only 3.5-4.0% of the size of the benzene peak in our sample of JP-4 (GEC-1A-792033).

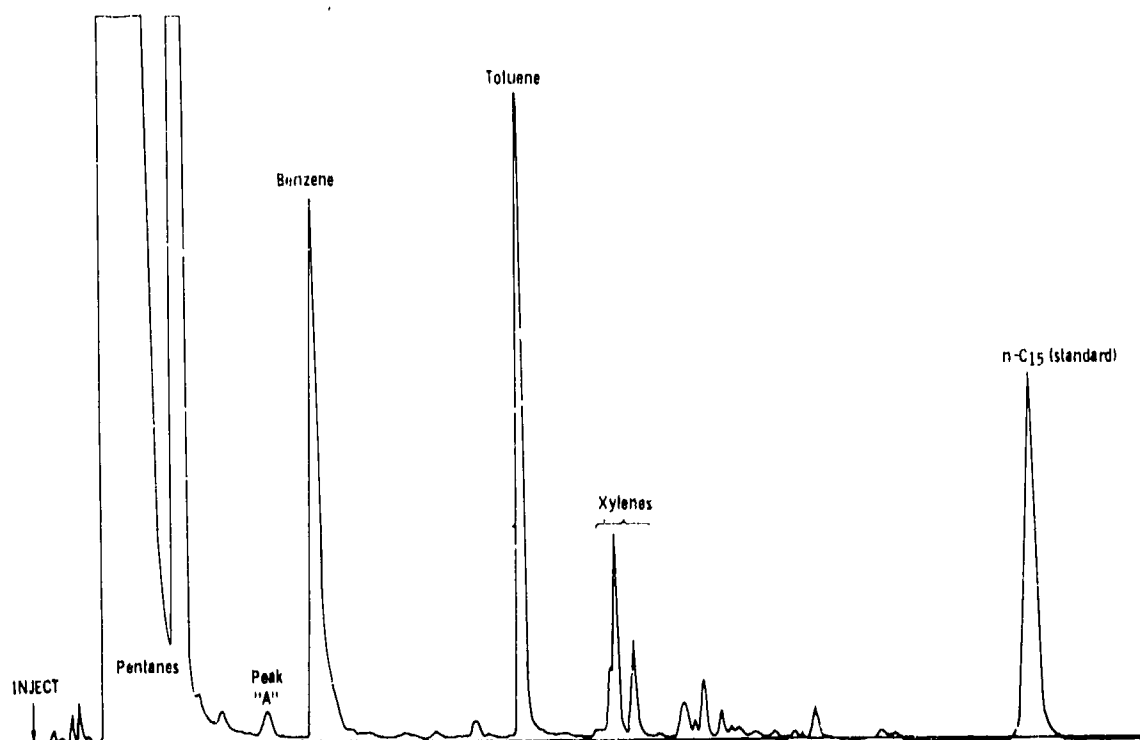


FIGURE 2. CHROMATOGRAM OF JP-4 WATER SOLUBLE EXTRACT

## PETROLEUM-BASED JP-4

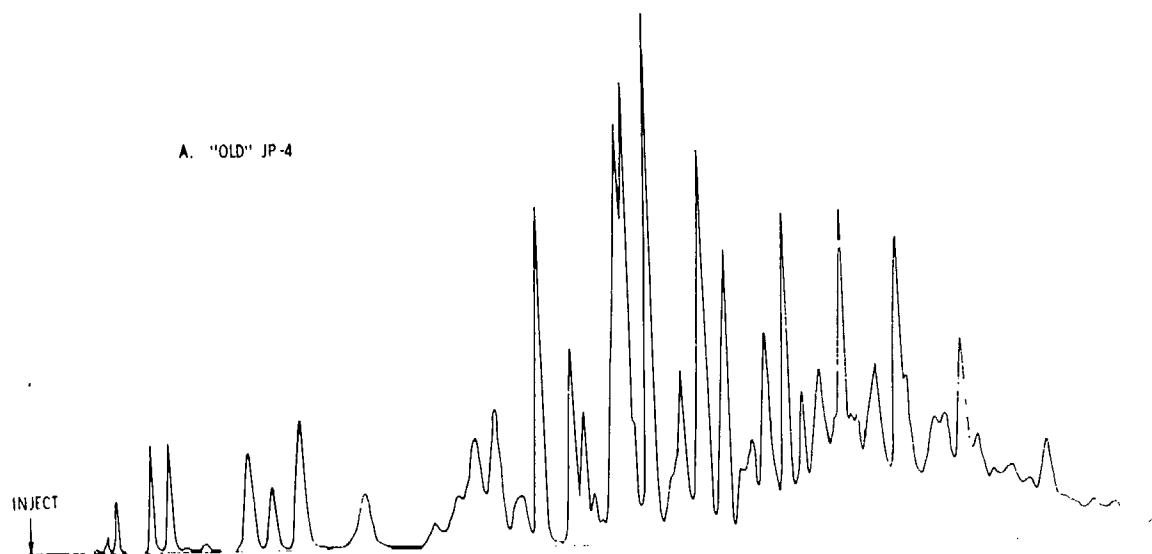
### Comparison of Neat JP-4 Samples

Since jet fuels, and JP-4 in particular, are supplied to a performance specification, it is evident that such a specification could be met by a wide range of fuel component combinations. Furthermore, since it is unlikely that the proportion of toxic components of (presently) unknown composition will fortuitously be constant, we have advocated the examination of a reasonable number (5 or 6) of different JP-4 samples. Unfortunately, to date these have not been forthcoming. During the current year we have received one 5-gallon sample each of a petroleum and a shale-based JP-4. In the absence of additional new JP-4 samples, we have tried to enlarge our sample group by including: a) our "old" (by several years) JP-4 Batch 2-13-79, b) a small sample of petroleum JP-4 supplied by Scherfig (U.C. Irvine), and c) a second small sample also supplied by Scherfig and stated to be the base JP-4, free of additives. We planned to use our gas chromatographic procedure to see how similar or different these JP-4 samples were.

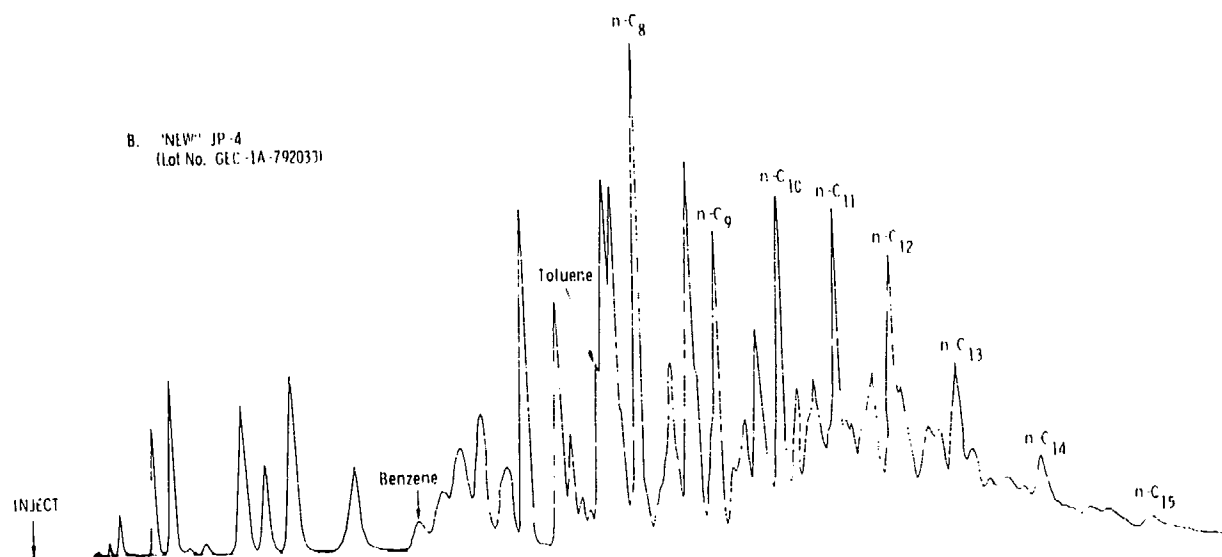
Results and Discussion. The gas chromatograms for each JP-4 sample are shown in Figure 3. Comparison of the chromatograms leads to the following conclusions.

1. The component compositions, at least for all the component peaks resolved in our chromatograms, are remarkably similar. That this is true even for JP-4 from such widely different sources as the petroleum and shale derived samples is shown by their superimposed chromatograms in Figure 4. Despite wide differences in relative amounts, most of the peaks match up.
2. As anticipated, differences can be seen in the relative amounts of the various component hydrocarbons in different samples. This of course emphasizes the need for a comparison of a range of samples before any general conclusions can be drawn regarding the overall toxicity of JP-4 fuels in the aquatic environment.
3. "Old" (Batch 3-13-79) and "new" (GEC-1A-792033) JP-4 samples: These two samples appear to be so similar even with regard to relative peak heights of virtually all the components that we conclude that they are probably part of the same batch, despite the large time interval between their dates of receipt (Note: we have been unable to find a batch identification number for the "old" sample but it has been used in the earlier bioassay work at SEEHRL (formerly SERL) for at least 2-3 years.)
4. JP-4 ex-Scherfig: Comparison of the chromatograms indicates that this sample is virtually identical to our "new" JP-4 (GEC-1A-792033) even to the relative peak areas. Since there are slight relative area differences around the known positions for the aromatics, benzene, toluene, and possibly xylene, it is possible that the two JP-4 samples were formulated from the same base fuel but with the addition of similar but not identical mixtures of aromatics.

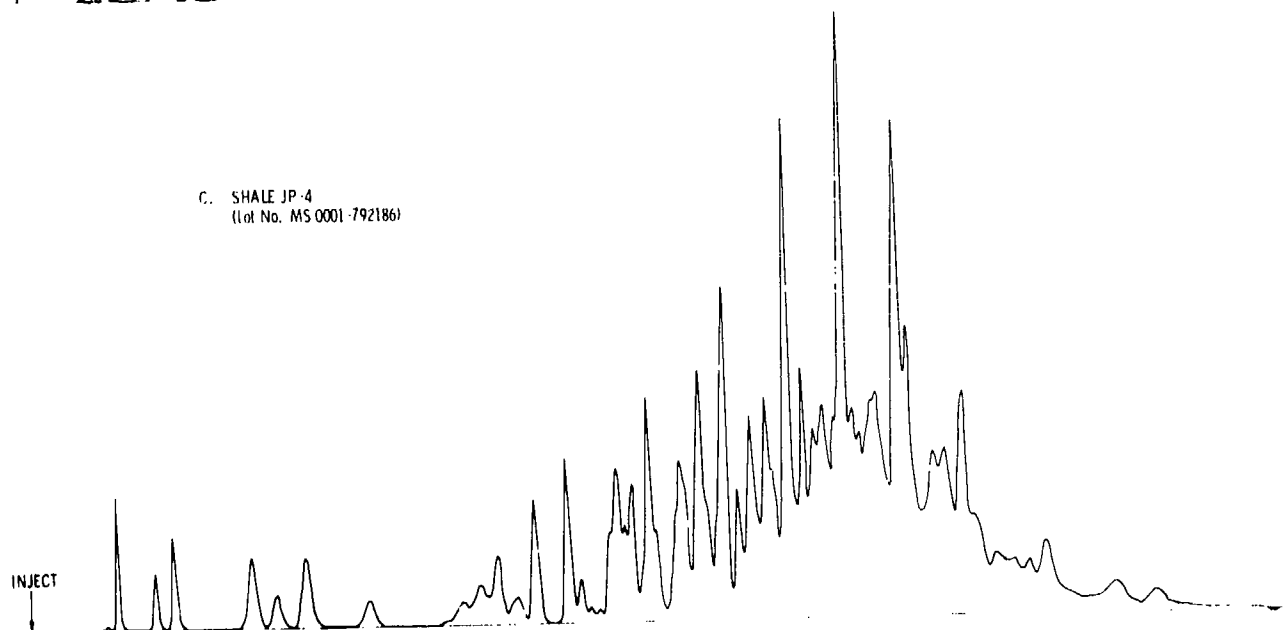
A. "OLD" JP-4



B. "NEW" JP-4  
(Lot No. GLC-1A-792037)



C. SHALE JP-4  
(Lot No. MS 0001-792186)



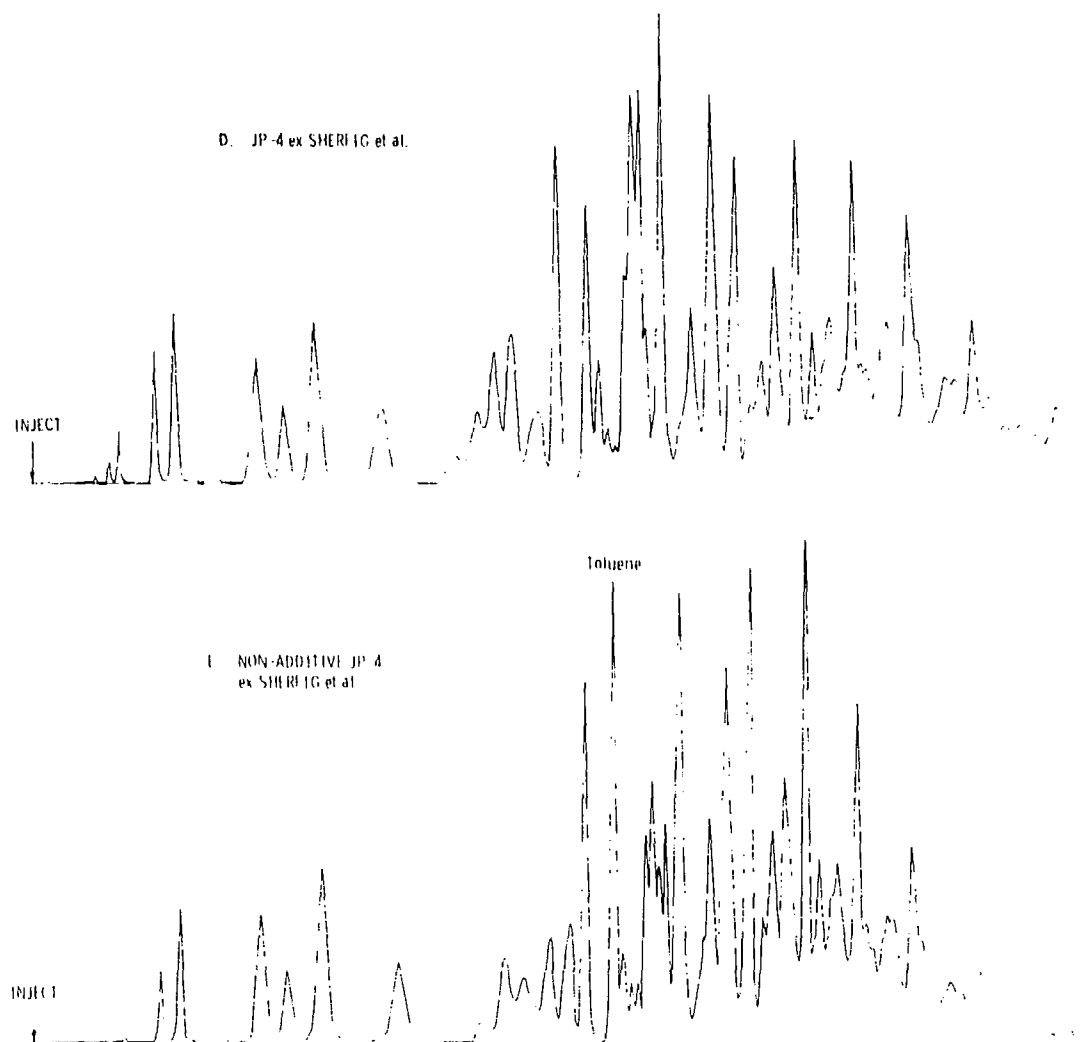


FIGURE 3. CHROMATOGRAMS OF JP-4 SAMPLES

- A. "Old" JP-4
- B. "New" JP-4 (Batch No. GEC-1A-792033)
- C. Shale JP-4 (Batch No. MS 0001-792186)
- D. JP-4 ex Scherfig
- E. "Non-additive" JP-4 ex Scherfig

5. "Additive-free" JP-4 ex-Scherfig: Again this JP-4 has a similar component composition to our "new" JP-4 and to Scherfig's JP-4 discussed above. However, the relative peak areas are quite different in many cases. Although this might be partly due to the absence of the "additives" the number of differences and their type, e.g. the different pattern of the major component n-alkanes in the C<sub>8</sub>-C<sub>14</sub> range, make it unlikely that this is the only explanation. Therefore, it must be concluded that this JP-4 base fuel comes from a different lot from our and Scherfig's whole fuels.
6. It is concluded that gas chromatography can readily be used to detect similarities and differences in different samples of JP-4 (and their water soluble extracts - see below). Furthermore, the differences detected in the few available samples described above are sufficiently great to reinforce the argument for examination of a wider range of JP-4 samples from different sources.

#### Water Soluble Extracts (WSF)

Preparation. (a) For Early *Daphnia* and *Artemia* Bioassay Tests: For these early tests, carried out before the preparative technique described under b) was developed, the WSF was prepared by vigorously shaking a 1:12 mixture of the fuel and water for approximately 2 min and allowing to separate and settle at least 24 h before withdrawing the aqueous phase for testing. Subsequently, when our analytical procedure had been developed an analysis (see Table 15) of a typical WSF of this type gave low values for hydrocarbon levels compared with procedure b) below. Thus it was concluded that the period of vigorous contact between the two phases was too short to ensure complete equilibration. (b) Preparation of True Equilibrated Water Extracts: Difficulties have been encountered in the preparation of true solutions of jet fuel hydrocarbons in water due to the ready formation of emulsified fuel droplets in the aqueous phase when water and fuel layers are mixed. While we recognize that in a real-life situation these fuel emulsions may be an inevitable part of the fuel/water system, it is essential at the outset to test the toxicity of truly dissolved hydrocarbons so that the toxic effects of the two systems (solution and emulsion) can be distinguished, especially since their mechanism of action may be quite different.

Other workers (Scherfig, 1981 and Laughlin et al., 1981) have reported on the difficulty of obtaining emulsion-free solutions. Laughlin has described a simple procedure involving the gentle contacting of water and fuel layers by magnetic stirring of the lower (water) phase at a rate slow enough to avoid vortexing. We have used this procedure to prepare consistently emulsion-free water extracts.

Water and fuel layers were contacted in aspirator bottles with a (lower) draw-off point in the water layer. The mixing was carried out in a constant temperature incubator. This avoided temperature swings that might cause the formation of fuel emulsion droplets in the aqueous phase. The temperature (20°C) was deliberately set below ambient so that withdrawal of the aqueous phase and transfer to room temperature conditions would not result in separation of a hydrocarbon phase. Gas chromatographic monitoring as described in an earlier section (page 22) was used to ensure the absence of

TABLE 15

## COMPOSITION OF WATER SOLUBLE EXTRACT FROM JP-4 (GEC-1A-792037)

	ppm "C <sub>15</sub> Units," $\pm 1$ SD	%
Benzene	$12.6 \pm 1.2$ (7.7) <sup>a</sup>	43
Toluene	$8.5 \pm 0.6$ (4.4)	29
Xylenes	$4.0 \pm 0.3$ (2.4)	14
Peak "A" (Alkane)	$0.47 \pm 0.06$ ( - )	2
Others (by difference)	- - -	12
Total Hydrocarbon	$29.3 \pm 2.2$ (17.9)	100
<hr/>		
Peak A / Benzene X 100	$4.0 \% \pm 0.3$	

<sup>a</sup> Typical values for the incompletely equilibrated WSF prepared by earlier procedure (see page 27 ) included for comparison.

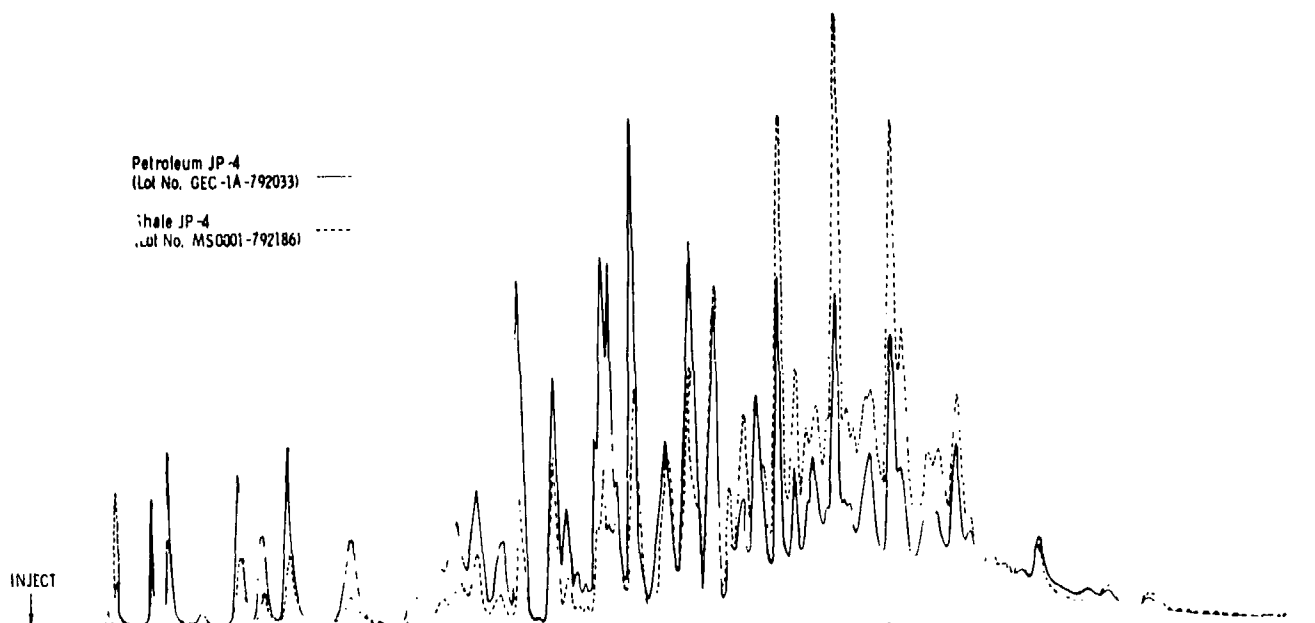


FIGURE 4. COMPARISON OF PETROLEUM-DERIVED AND SHALE-DERIVED JP-4 SAMPLES

any substantial amount of fuel emulsion in the aqueous phase.

Monitoring of WSF Dilutions for Bioassays. In the past there has been no check by actual analysis on the WSF dilutions used in our bioassay tests. Such a check is now seen to be especially desirable in view of the loss of hydrocarbons known to take place during storage in glass bottles (see below). Our chromatographic procedure makes it possible to monitor not only the levels of total hydrocarbons but also of any of the major components of the water soluble extract. A typical analysis (Table 16) of a series of WSF dilutions shows the generally excellent agreement between calculated and measured dilutions.

TABLE 16  
COMPARISON OF CALCULATED AND MEASURED WSF DILUTIONS

Calculated:	% of Original WSF						
	100	65	42	32	18	6.5	3.2
Measured:							
Based on:							
1. Total Hydrocarbons	100	69.5	45	32	18	5	1.7
2. Benzene	100	69	46	34	18	6	2.3
3. Toluene	100	67	46	32.5	18	5	1.8
4. Xylenes	100	69	47	35	22	5	1.1

Composition. A typical water soluble extract is shown in Figure 2. Based on analyses of seven different extracts, an average composition was determined (Table 15).

It will be seen that the three simplest aromatic hydrocarbons - benzene, toluene, and xylene (isomers) - account for almost 90% of the water soluble hydrocarbons in this sample of JP-4 jet fuel.

Factors Affecting Preparation. (a) Fuel/Water Ratio: For reproducibility in testing it is obviously desirable to work with a water soluble extract that is saturated or equilibrated at a given temperature in all the hydrocarbon components extracted from the JP-4 fuel. Thus it is desirable to obtain some idea of suitable fuel/water ratios for producing this kind of extract.

Samples of water soluble extracts were prepared simultaneously at two fuel/H<sub>2</sub>O ratios, 1:12 and 1:6, and at the same constant temperature (20°C). Analysis by chromatography gave the following results (Table 17).

These results agree within the reproducibility (SD  $\approx$  3-5%) of our analytical procedure, indicating that a ratio of 1:12 is adequate for preparing the water soluble extract.



TABLE 17  
EFFECT OF FUEL/H<sub>2</sub>O RATIO ON COMPOSITION  
OF WATER SOLUBLES FROM JP-4

	Concentration, ppm "C <sub>15</sub> Units"	
	1:12	1:6
Benzene	13.8	14.2
Toluene	9.1	9.5
Xylenes	4.4	4.5
Total Hydrocarbons	31.5	33.8

(b) Fuel/H<sub>2</sub>O Contact Time: In view of the very gentle contacting technique necessary to prevent the introduction of fuel emulsion droplets into the water phase, it was felt that extended contact periods might be necessary to establish true equilibrium (saturation).

Water extractions, using 1:12 fuel/H<sub>2</sub>O, were carried out with 24-h, 48-h, and 72-h contact periods. The water phases gave the following analyses (Table 18).

TABLE 18  
EFFECT OF FUEL/H<sub>2</sub>O CONTACT TIME ON COMPOSITION  
OF WATER SOLUBLES

	Concentration, ppm "C <sub>15</sub> Units"		
	24-h	48-h	72-h
Benzene	13.5, 14.1	14.4	12.6
Toluene	8.9, 9.4	9.7	8.5
Xylenes	4.1, 4.5	4.7	4.0
Total Hydrocarbons	29.7, 31.6	32.5	29.7

All the runs agree within our reproducibility, indicating that equilibration is essentially achieved within 24 h.

Factors Affecting Use in Bioassays. In assessing the toxicity of the WSF components it is essential to ensure the consistency of the WSF both from test to test and especially within the period (1-3 days) of each test. This is especially important in view of the low levels (ppm) of test hydrocarbons since what would normally be considered minor losses or contaminant pickup may represent a substantial fraction of the test components. The

following series of experiments were designed to assess the magnitude of potential hydrocarbon losses during bioassay runs.

(a) Effect of Storage Conditions: Samples of JP-4 water soluble extract, prepared as described (page 27) were stored in glass bottles under a variety of conditions designed to explore: a) the extent of biodegradation (by storing with and without the addition of  $\text{HgCl}_2$ , b) the effect of full and partially filled bottles, and c) the effect of silanizing the glass bottle surfaces. Chromatography analysis of controls and the stored samples gave the results summarized in Tables 19 and 20 in terms of hydrocarbon loss.

TABLE 19  
EFFECT OF  $\text{HgCl}_2$  ON STORED SAMPLES  
Storage Period: 48 h

	Loss, % <sup>a)</sup>	
	<u><math>\text{HgCl}_2</math></u>	<u>No <math>\text{HgCl}_2</math></u>
Benzene	29	33
Toluene	24	29
Xylenes	20	31
Peak "A"	94	100
Total Hydrocarbons	28	34

<sup>a</sup> Relative to control sample stored for 0 h.

TABLE 20  
EFFECT OF STORAGE IN FULL, PARTIALLY FILLED,  
AND SILANIZED BOTTLES<sup>a</sup>  
Storage Period: 6 h

	Loss, % <sup>b)</sup>		
	<u>Partially Full</u>	<u>Full</u>	<u>Silanized</u>
Benzene	23	20	32
Toluene	23	NM <sup>d</sup>	28
Xylenes	NM <sup>c</sup>	26	31
Peak "A"	42	21 <sup>d</sup>	45
Total Hydrocarbons	23	NM <sup>d</sup>	31

<sup>a</sup> Results not comparable with Table 19; different sized bottles used.

<sup>b</sup> Relative to controls stored for 0 h.

<sup>c</sup> Not measured.

<sup>d</sup> Not measured due to contaminant emerging under toluene peak.

All samples tested showed substantial losses of all the hydrocarbons measured. Since the test for biodegradation showed at most a minor increase in hydrocarbon loss, even in the absence of the antibacterial  $\text{HgCl}_2$  (Table 19) the bulk of the loss appears to be due to surface adsorption by the glass or vaporization into any air space within the bottles. The comparison of filled and partially filled containers in Table 20 indicates that the presence or absence of air space has little effect on the loss of the major hydrocarbons. Thus surface adsorption appears to be the primary mechanism responsible for the disappearance of the hydrocarbons.

Silanization of the glass surfaces was of interest since it might react with "active sites" and deactivate the surface. If anything, the opposite effect was observed (Table 20). The apparent increase in the hydrocarbon loss is probably due to increased attraction of the bonded alkyl groups on the silanized surface for the water soluble hydrocarbons.

(b) Effect of Storage Time: The previous section showed the persistent loss of hydrocarbons from the water soluble extract under a variety of storage conditions. Since bioassay procedures generally require at least a 1-3 day test period, it was important to assess the extent of hydrocarbon losses over such extended periods.

Water soluble extracts were stored at  $20^\circ\text{C}$  for up to 48 h in a series of bottles, some filled to the top, some partially filled, and finally some pre-silanized. Samples were analyzed after 1, 6, 24, and 48 h. Losses relative to (unstored) controls were calculated (Tables 21 and 22) and plotted in Figure 5. It was seen that:

1. Hydrocarbon losses were confirmed under all conditions.
2. Using nonsilanized bottles, losses of total hydrocarbons and aromatics ranged as high as 60-70% before leveling off (presumably as the glass surface becomes saturated).
3. Losses of the minor peak "A", presumably an alkane since it emerges just prior to benzene, went much higher (80-90%) both in untreated and silanized bottles. Since it was unlikely that saturated hydrocarbons were adsorbed to the glass more strongly than the more polar aromatics, it is probable that most of this loss is azeotropic evaporation into the air space in the bottle. This hypothesis appears to be confirmed by the additional observation that in tests with a completely filled bottle (i.e., no air space) the peak "A" losses dropped to the same level as the aromatics.
4. With silanized bottles overall loss of aromatics (and total hydrocarbons) was markedly reduced (to ~ 30-40%), presumably as a result of reduction in the number or the activity of adsorption sites on the glass surface. At the same time the more rapid uptake by the glass, previously noted (Table 20), was confirmed. Thus in the absence of any other alternative (but see below) silanization treatment of bioassay test bottles would appear to offer advantages with regard to reduced hydrocarbon losses and more rapid equilibration.

TABLE 21

EFFECT OF STORAGE TIME ON HYDROCARBON LOSSES -  
UNSILANIZED BOTTLES

Storage Time, h:	Loss, %			
	<u>1</u>	<u>6</u>	<u>24</u>	<u>48</u>
Benzene	2	23	61	69
Toluene	4	23	54	65
Peak "A"	10	42	NM <sup>a</sup>	91
Total Hydrocarbon	7	23	60	68

<sup>a</sup> Not measured.

TABLE 22

EFFECT OF STORAGE TIME ON HYDROCARBON LOSSES -  
SILANIZED BOTTLES

Storage Time, h:	Loss, %		
	<u>1</u>	<u>6</u>	<u>24</u>
Benzene	24	32	39
Toluene	19	28	37
Peak "A"	29	45	82
Total Hydrocarbon	21	31	40

(c) Effect of Pretreatment with Water Soluble Extract: The loss of hydrocarbons from JP-4 water soluble extracts in glass containers introduces a substantial source of error into bioassays using this test material. If our hypothesis of adsorption onto the glass surface is correct, it should be possible to pre-equilibrate the container with a sacrificial portion of the water extract. Losses from a second portion of the extract should then be minimized. This was confirmed in a preliminary series of tests with water soluble extract dilutions that were also being used to evaluate the feasibility of *Artemia* egg hatchability as a bioassay technique. A portion of each dilution was used to treat the assay bottles for 24 h before the actual test sample was substituted. The samples were then stored at 25°C for the period of the hatchability tests (66 h) at which time 50-ml samples were analyzed. Representative data are tabulated below (Table 23).

These results show a substantial reduction in hydrocarbon losses since our earlier tests would have predicted losses of 60-70% or greater from a 66-h storage period. Furthermore, it should be possible to eliminate losses completely by pre-saturating with a more concentrated water extract than the test sample.

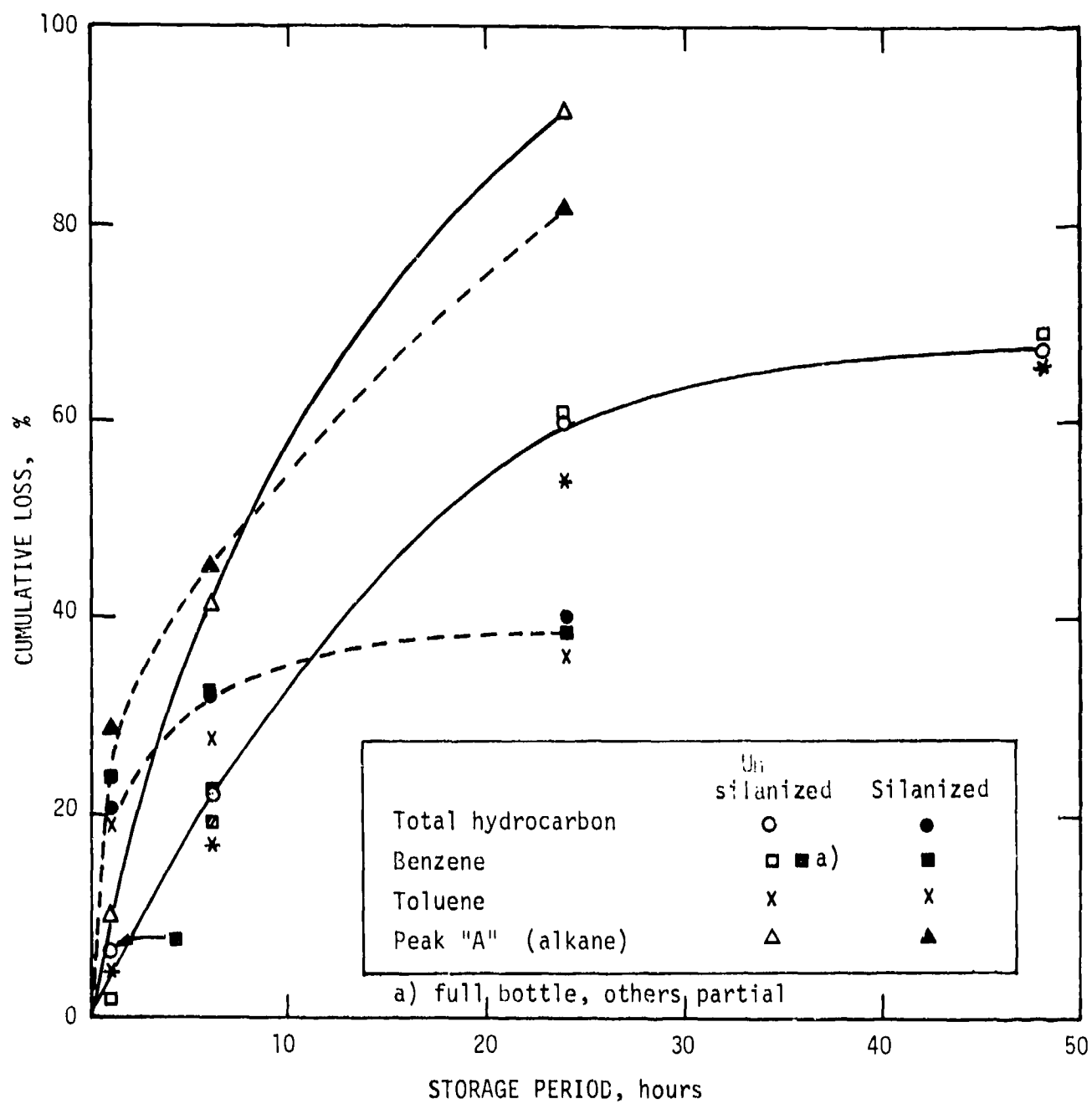


FIGURE 5. EFFECT OF STORAGE IN GLASS (SILANIZED AND UNTREATED) ON JP-4 WATER SOLUBLE EXTRACT HYDROCARBONS

TABLE 23  
HYDROCARBON LOSSES AFTER STORAGE FOR  
66 HOURS IN PRETREATED BOTTLES

	Loss, %		
	65 % Dilution	42 % Dilution	32 % Dilution
Total Hydrocarbon	18	21	16
Benzene	18	22	15
Toluene	14	23	14
Xylenes	11	20	18
Peak "A" (Alkanes)	22	32	18

#### SHALE-BASED JP-4

##### Neat Fuel - Comparison with Petroleum-Based JP-4

As mentioned earlier (Figure 4), the shale JP-4 (Batch No. MS 0001-792186) appears to have similar component composition to our "new" JP-4 sample (GEC-1A-792033). Nevertheless, there are substantial differences in relative peak areas, including the following:

- (a) Total peak area is 12% less for the same-size sample injection.

Shale JP-4	447193 area units
Petroleum JP-4	505231 area units

This difference is much greater than our percent standard deviation for successive injections of the same JP-4 sample (<3%).

- (b) One major difference appears to be a smaller proportion of lower boiling components, represented by the first half of the chromatogram, in the shale JP-4.

		Percentage of Total Peak Area
Peaks in 1st Half:	Shale JP-4	37
	Petroleum JP-4	46

- (c) The one exception to this decrease in volatile components is the very volatile third peak which is 6 times greater in the shale sample. It is possible that this component (probably a C<sub>4</sub> or C<sub>5</sub> hydrocarbon) has been added to improve the volatility characteristics of the shale JP-4.

## Water Soluble Extracts (WSF)

Preparation. Water soluble extracts were prepared in similar fashion to the petroleum-based JP-4.

Composition - Comparison with Petroleum JP-4 Extract. Figure 6 shows a typical chromatogram of the water extract. The major component composition of the shale extract is summarized in Table 24 together with a similar listing for the petroleum-based fuel. There are substantial differences between the extracts including:

1. Substantially reduced levels of all tabulated components in the shale extract.
2. The three aromatics, benzene, toluene, and xylenes, still comprise the major portion of the water solubles but at a reduced level (70% of total compared with 86%).
3. The reduction in total hydrocarbons is accounted for by the overall drop in aromatics. In other words, alkane levels are about the same. This means that the reduction in water solubles is probably due mainly to lower concentrations of aromatics in the neat shale jet fuel, estimated as follows:

Benzene	80 % less
Toluene	40 % less
Xylenes	15 % less

In addition to these differences among the major components of the water soluble extract, comparison of the high-boiling minor components emerging at the end of the chromatogram reveal a number of differences (compare Figures 6 and 5).

TABLE 24  
COMPARISON OF WATER EXTRACTS FROM SHALE AND  
PETROLEUM-DERIVED JP-4

	Conc., ppm w/v "C <sub>15</sub> Units"		$\Delta$ % (Petroleum-Shale)
	Shale JP-4	Petroleum JP-4	
Total Hydrocarbons	15.8	29.3	46
Benzene	2.7	12.6	70
Toluene	5.1	8.5	40
Xylenes	3.4	4.0	15
Peak "A" (Alkane)	0.21	0.47	55
<hr/>			
Peak A / Benzene X 100	7.7	4.0	

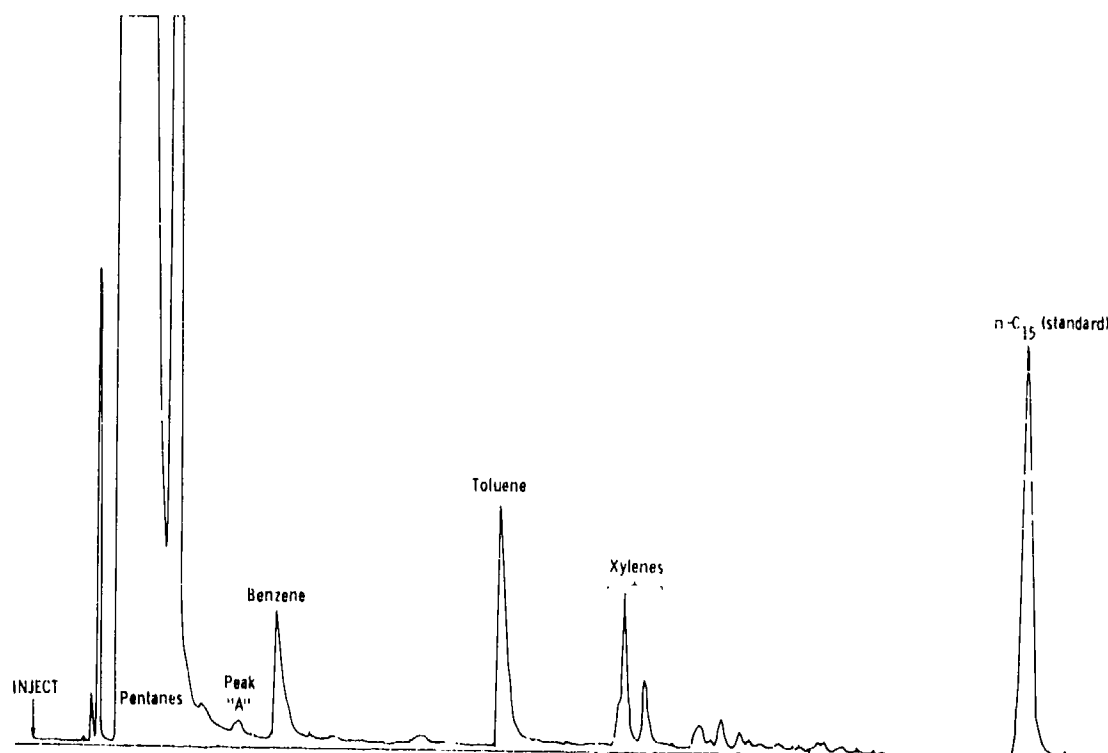


FIGURE 6. CHROMATOGRAM OF SHALE JP-4 WATER SOLUBLE EXTRACT

Factors Affecting Preparation. Evaluation of the effects of different fuel/H<sub>2</sub>O ratios and holding times prior to sampling, summarized in Table 25,

TABLE 25  
EFFECT OF FUEL/H<sub>2</sub>O RATIOS AND HOLD TIMES  
ON SHALE JP-4 EXTRACT

	Conc., ppm w/v "C <sub>15</sub> Units"		
	Fuel/H <sub>2</sub> O = 1/6		Fuel/H <sub>2</sub> O = 1/12
	0-h Hold	0-h Hold	24-h Hold
Total Hydrocarbons	16.8	15.7	15.0
Benzene	2.9	2.7	2.6
Toluene	5.5	4.95	4.8
Xylenes	3.5	3.3	3.3
Peak "A"	0.21	0.21	0.22

confirm the observations on petroleum JP-4 extract, namely that a 1:12 fuel/H<sub>2</sub>O ratio is sufficient to produce an equilibrated extract and that the extract can be used immediately after the mixing period (without holding for 24 h) provided that careful, gentle nonvortexing mixing has been maintained.



## MATHEMATICAL MODELING

### METHODOLOGY

Mathematical modeling is an activity which can take quite different forms depending on data available and on the objective to be met by the final product. In the restricted area of mathematical modeling as it applies to aquatic systems there are at least four schools of thought: simulation modellers vs. proponents of "black box" or principally data-based models and, on a different dimension, the hydrologic modellers vs. those more concerned with water quality. There are obviously combinations of these approaches which can be and are applied to particular problems but to most practitioners a modeling study conjures up the particular approach and school of thought with which they are most familiar. As a consequence it is important here to make clear the approach being taken to the fuel spill problem because it differs from most of the conventional procedures both in spirit and in object.

At the stage of the investigation with which we are here concerned, there is no alternative to utilizing some type of simulation model as the mathematical format into which assumptions regarding causal relations and parameter values are summarized. By simulation model we mean one whose structure and parameters are explicitly related to physical, chemical, or biological processes. Data in the literature on algal growth rates as a function of nutrient level, for example, are often given in terms of Michaelis constants, a fact which points out that simulation models are constrained to be written in the language of the various disciplines which have studied the component processes of the system. This constraint immediately leads to the result that most simulation models will be complex with many parameters, state variables, and nonlinear relations. Under the best circumstances, such models have many degrees of freedom and, with judicious manipulation, can be made to produce virtually any desired behavior, often with both plausible structure and parameter values. Because of this problem, simulation modeling has limited importance in cases where extensive data sets that quantify the system behavior are lacking.

In spite of the cited problems, the potential utility of information yielded by simulation models in planning experiments has been recognized. For example, with reference to ecological models, Jeffers (1972) states that:

"Much time can be saved in the early stages of hypothesis formulation by the exploration of these hypotheses through mathematical models. Similarly mathematical models can be used readily to investigate phenomena from the viewpoint of existing theories, by the integration of disparate theories into a single working hypothesis, for example. Such models may quickly reveal inadequacies in the current theory and indicate gaps where new theory is required."

Similarly, Mar (1974) in his review of multidisciplinary modeling studies pointed out that:

"The strategy to construct models without data and then employ sensitivity analysis to identify critical components where research and new data would enhance model performance is not commonly practiced."

Stenseth (1977), while roundly criticizing simulation modeling, admits that a simple model when used to explore or to generate hypotheses can be a valuable research tool. Unfortunately, little work has been done to elucidate exactly how mathematical models can actually be used in such a "Hypotheses generating" mode.

Several workers (e.g. Adams, 1972; Meyer, 1972; Maddock, 1973; McCuen, 1976) have suggested that parameter sensitivity analysis can be used to guide future data collection efforts and/or to order research priorities. Traditional parameter sensitivity analysis, however, pertains to a particular point in the parameter space (the vector space spanned by all possible combinations of parameter values). This requires that point estimates of all parameters be available, which, in turn, for complex environmental models, implies that sufficient input-output data for model calibration exist, and this is counter to our original premise that application was to be for an early stage of the investigation. Although Meyer (1972) advocates the use of a "tentatively calibrated model" to overcome this particular dilemma, the structure of models of environmental systems is not likely to be well defined a priori, and such an approach is therefore suspect.

In light of the discussion above, we contend that, in the early stages of the analysis of real problems, simulation models can be useful only in a probabilistic context. This is, given the model and the inherent uncertainties in structure and parameter values, the only meaningful analysis must focus on the probabilities of various behaviors. Most importantly, it must focus on the probable structures and parametric relations which appear consistent with that behavior which is associated with the "environmental problem" under consideration. One method for applying simulation models in a probabilistic context is to use Monte Carlo techniques. (For example, see Tiwari and Hobbie (1976) and Tiwari et al. (1978) for an application of Monte Carlo simulation in ecological modeling.) The methodology developed below adjoins the notion of qualitative or semi-quantitative descriptors of the behavior of the system to Monte Carlo simulation to obtain a useful technique for the preliminary analysis of environmental systems.

For clarity of exposition, we restrict our attention to a specific class of models and introduce nomenclature which will be required subsequently. Assume the processes are to be modeled by a set of first order ordinary differential equations. (Different mathematical structures can be dealt with in an analogous way.) Let these equations be given in the form:

$$\frac{dx(t)}{dt} = \dot{x}(t) = f[x(t), \xi, z(t)] \quad (1)$$

where  $x(t)$  is the state vector and  $z(t)$  a set of time variable functions which include input or forcing functions. The vector  $\xi$  is a set of constant parameters described more fully below. Thus, for  $\xi$ ,  $z(t)$ , and  $x(0)$

specified,  $x(t)$  is the solution of the system of equations and is a deterministic or stochastic function of time as determined by the nature of  $z(t)$ . For simplicity of exposition,  $z(t)$  will be treated hereafter as a deterministic function of  $t$ . Under this assumption, there are two types of uncertainty with which we will deal: uncertainty in the model structure, i.e. in the functions,  $f$ , and uncertainty in the parameter values,  $\xi$ . Different model structures would pertain to competing hypotheses on system functioning (e.g. phosphorus limitation versus nitrogen limitation in a eutrophication problem); we use the term scenario to indicate a particular structure.

For a given scenario, each element of the vector  $\xi$  is defined as a random variable, the distribution of which is a measure of our uncertainty in the "real" but unknown value of the parameter. These parameter distributions are formed from data available from the literature and from experience with similar structures. For example, the literature suggests that the maximum growth rate of Chlorella vulgaris is almost certainly between 1.5 and 2.5 days<sup>-1</sup> at water temperatures near 25°C. Interpreting these limits as the range of a rectangularly distributed random variable, and forming similar a priori estimates for the other elements of  $\xi$  result in the definition of an ensemble of models for a given scenario. Some of these models will, we hope, mimic the real system with respect to the behavior of interest.

Turning now to the question of behavior, recall that, for a given scenario, every sample value of  $\xi$ , drawn from the a priori distribution, results in a unique state trajectory,  $x(t)$ . Following the usual practice, we assume that there is a set of observed variables  $y(t)$ , calculable from the state vector, which is important to the problem at hand. So, for each randomly chosen parameter set  $\xi$  there corresponds a unique observation vector  $y(t)$ . Since the elements of  $y(t)$  are observed (that is we assume that they are measured in the real system), it is sensible to define behavior in terms of  $y(t)$ . For example, suppose  $y_i$  is the concentration of phytoplankton in a body of water and the problem in question concerns unwanted algal blooms due to nutrient enrichment. Then, there is some value of  $y_i$  above which a bloom is defined to have occurred and the behavior is defined by this critical value.

In general, a number of behavior categories can be used. Without loss of generality, however, we can consider the case for which behavior is defined in a binary sense, i.e., it either occurs or does not occur for a given scenario and set of parameters  $\xi$ . It follows that a rule must be specified for determining the occurrence or non-occurrence of the behavior on the basis of the pattern of  $y(t)$ . It is also possible that the behavior might depend on the vector  $z(t)$ . For example, suppose one element of  $z(t)$  was water temperature. We might be interested only in extreme values of  $y(t)$  when adjusted or controlled for temperature variations. In any event, the detailed definition of behavior is problem-dependent and, for present purposes, it is sufficient to keep in mind that a set of numerical values of  $\xi$  leads to a unique time function  $y(t)$  which, in turn, determines the occurrence or non-occurrence of the behavior conditioned, perhaps, by  $z(t)$ .

We have now presented the class of models to be studied, defined the scenario concept and described how we propose to deal with parametric

uncertainty. For a given scenario, behavior and set of parameter distributions  $\xi$ , it is possible to explore the properties of the ensemble via computer simulation studies. In particular, a random choice of the parameter vector  $\xi$  from the predefined distributions leads to a state trajectory  $x(t)$ , an observation vector  $y(t)$  and, via the behavior-defining algorithm, to a determination of the occurrence of the behavior. A repetition of this process for many sets of randomly chosen parameters results in a set of sample parameter vectors for which the behavior was observed and a set for which the behavior was not observed. The key idea is then to attempt to identify the subset of physically, chemically, or biologically meaningful parameters which appear to account for the occurrence or non-occurrence of the behavior. More traditional sensitivity analyses of large ecological models inevitably show that a surprisingly large fraction of the total number of parameters is simply unimportant to the critical model behavior. We maintain that this unimportant subset, or, conversely, the critical subset, may be tentatively specified rather early in any study.

Ranking the elements of  $\xi$  in order of importance in the behavioral context is accomplished through an analysis of the Monte Carlo results. The essential concept can best be illustrated by considering a single element  $\xi_k$ , of the vector  $\xi$  and its a priori cumulative distribution, as shown in Figure 7. Recall that the procedure is to draw a random sample from this parent distribution (a similar procedure is followed for all other elements of  $\xi$ ), run the simulation with this value and record the observed behavior and the total vector  $\xi$  therewith associated. A repetition of this procedure results in two sets of values of  $\xi_k$ , one associated with the occurrence of the behavior B, the other without the behavior  $\bar{B}$ . That is, we have split the distribution  $F(\xi_k)$  into two parts as indicated in the figure. This particular example would suggest that  $\xi_k$  was important to the behavior since  $F(\xi_k)$  is clearly divided by the behavioral classification. Alternatively, if the sample values under B and  $\bar{B}$  appeared both to be from the original distribution  $F(\xi_k)$ , then we would conclude that  $\xi_k$  was not important.

For the case where  $z(t)$  is a deterministic function of time, the parameter space is cleanly divided by the behavioral mapping; that is, there is no ambiguity regarding whether a given parameter vector results in B or  $\bar{B}$ . Our analysis then focuses on the determination of which parameters or combinations of parameters are most important in distinguishing between B and  $\bar{B}$ . We will restrict the discussion to the case for which the parameter vector mean is zero and the parameter covariance matrix is the identity matrix. (A suitable transformation can always be found to convert the general problem to this case.) The problem of identifying how the behavioral mapping separates the parent parameter space can then be approached by examining induced mean shifts and induced covariance structure.

For example, we can base a sensitivity ranking on a direct measure of the separation of the cumulative distribution functions,  $F(\xi_k|B)$  and  $F(\xi_k|\bar{B})$ . In particular, we utilize the statistic:

$$d_{m,n} = \sup_x S_n(x) - S_m(x)$$

where  $S_n$  and  $S_m$  are the sample distribution functions corresponding to  $\xi_k|B$  and  $F(\xi_k|\bar{B})$  for  $n$  behaviors and  $m$  non-behaviors. The statistic  $d_{m,n}$  is that used in the Kolmogorov-Smirnov two sample test and both its asymptotic and small sample distributions are known for any continuous cumulative distribution function  $F(\xi_k|B)$  and  $F(\xi_k|\bar{B})$ . Since  $S_n$  and  $S_m$  are estimates of  $F(\xi_k|B)$  and  $F(\xi_k|\bar{B})$ , we see that  $d_{m,n}$  is the maximum vertical distance between these two curves and the statistic is, therefore, sensitive not only to differences in central tendency but to any difference in the distribution functions. Thus, large values of  $d_{m,n}$  indicate that the parameter is important for simulating the behavior and, at least in cases where induced covariance is small, the converse is true for small values of that statistic.

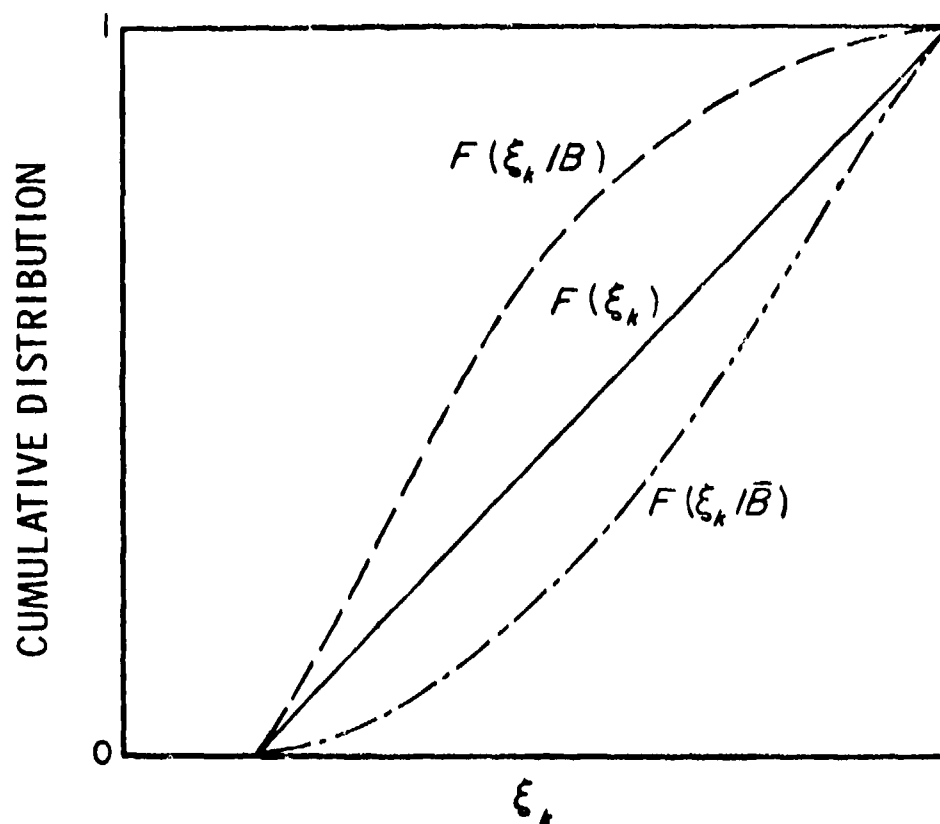


FIGURE 7. CUMULATIVE DISTRIBUTION FUNCTIONS FOR PARAMETER  $\xi_k$

$F(\xi_k)$  = parent, a priori distribution,  $F(\xi_k|B)$  = distribution of  $\xi_k$  in the behavior category,  
 $F(\xi_k|\bar{B})$  = distribution of  $\xi_k$  in the non-behavior category.

## APPLICATION TO THE JET FUEL PROBLEM

Given then, that our objective is to develop a simulation model to guide the collection and synthesis of data on the impact of jet fuel spills on aquatic ecosystems and, given further that the methodological approach is chosen, we may turn to a discussion of the properties of a model appropriate to these ends. Preliminary laboratory work indicates that the principal toxic constituents of these fuels are likely to be benzene, toluene, and xylene. There are essentially four "compartments" in the aquatic environment into which it seems sensible to partition these compounds: a slick floating on the water surface, a soluble fraction in the water column, a fraction suspended in the water column in the form of an emulsion, and a sediment compartment. Let us denote benzene by  $x$ , toluene by  $y$ , and xylene by  $z$ . Further, let the slick be compartment 1, the soluble fraction compartment 2, the emulsified fraction compartment 3, and the sediment compartment 4. Then, for each chemical constituent, we may denote the mass transfers in a lumped parameter differential equation model, at least in structural form, as:

$$\dot{x}_1 = -k_{11}x_1 + k_{12}x_2 + k_{13}x_3$$

$$\dot{x}_2 = k_{21}x_1 - k_{22}x_2 + k_{23}x_3 + k_{24}x_4$$

$$\dot{x}_3 = k_{31}x_1 + k_{32}x_2 - k_{33}x_3$$

$$\dot{x}_4 = k_{41}x_1 + k_{42}x_2 + k_{43}x_3 - k_{44}x_4$$

where  $\dot{x}_i = dx_i/dt$ . The form of the model is apparently linear, but any given term  $k_{ij}x_j$  is meant only to imply that there is a potential mass exchange between compartments  $i$  and  $j$ . The functional form we may expect to be much more complex. The form (or forms) of these terms will be dictated by the best information available in component disciplines involved. For example, Stolzenbach et al. (1977) reviewed the state of the art of mathematical modeling techniques for oil slick behaviors with respect to biodegradation, evaporation, and emulsification as well as transport by winds and currents. Since model components tend to be developed by scientists following a more-or-less reductionist approach, the form of the terms in the model are likely to be complex. Indeed, in general the  $k_{ij}$  terms must be regarded as being complicated functions of  $x$ ,  $y$ , and  $z$  as well as being dependent on the constituents of the fuel that are not accounted for in the model. That is, there is uncertainty inherent in this model or any other of such a complex environmental phenomenon.

To further elucidate the approach to the fuel spill problem, let us consider only the volatilization of  $x$ ,  $y$ , and  $z$  from the slick. The issue here is to predict the evaporative mass transfer to the vapor phase based on a knowledge of the composition of the fuel in the liquid phase in the slick. At a first cut this transfer rate depends on the various vapor pressures, the

activity coefficients, and a mass transfer coefficient. Popendorf (1981) has recently dealt with this problem in the context of mixed solvent vapor exposures in industrial hygiene using chemical thermodynamic methods. He suggests that reasonable predictions can be made in the present case if the water surface is treated as an infinite flat plate and if some data can be obtained on the solubility of water in the fuel. Moreover, it is possible to check such predictions in a straightforward manner using Popendorf's existing experimental set up. Hence, it appears to be feasible to characterize mathematically an important transfer process and to obtain realistic parameter bounds for this subprocess.

Similarly, it appears possible to characterize the transfer between the slick and the soluble fraction in the water column. Hunter (1981) has found, however, that the actual levels of  $x$ ,  $y$ , and  $z$  found in water solutions seldom approach their published saturation values. While this transfer from the slick to the water column will probably be rather tractable analytically, the relations between the slick, the emulsion compartment, and the soluble fraction must be regarded as much more difficult. Hunter also reports that it takes very little energy input to a separated water/fuel mixture to result in an emulsion in the laboratory setting. Clearly, the particle size distribution of the emulsion will be an important variable which mediates transfer between the emulsion and the soluble fraction. Further, the dynamic state of the slick/water interface will be central to understanding the development and persistence of the emulsion. There is a substantial literature on these issues particularly in chemical engineering. Further, the emulsion issue clearly indicates the first benefit of the modeling approach: it is almost impossible to attack the overall problem without a very thorough compilation and synthesis of the physical chemistry and mass transport aspects of the problem.

In general, it will be necessary to deal with each transfer rate in similar detail. At this very early stage we speculate that the air/slick transfer is tractable as is the sediment/soluble fraction transfer. The key is, as indicated above, the emulsion issue. It seems likely that the emulsion issue will be central to the ultimate problem, that of predicting toxicity to the aquatic community. We may expect there to be species within any community that will be principally exposed to the toxic agents  $x$ ,  $y$ , and  $z$  that are dissolved in the water column. Conversely, species higher on the food chain may be principally exposed to the emulsion. If  $y_k$  denotes the toxicity of the fuel to the  $k$ -th species the toxicity function can be supposed to have the form:

$$y_k = f_k(x_2, y_2, z_2) + g_k(x_3, y_3, z_3)$$

where  $f_k$  is the toxicity of the soluble fraction and  $g_k$  that of the emulsified fraction. If there is no synergistic toxicity then, to a first approximation

$$y_k = c_{1k}x_2 + c_{2k}y_2 + c_{3k}z_2 + c_{4k}x_3 + c_{5k}y_3 + c_{6k}z_3$$

where

$$c_{1k} = \frac{df_k}{dx_2} \quad \text{etc.}$$

The toxicity index raises two issues, that of defining the species impacted and that pertaining to acute or chronic toxicity. That is, immediately subsequent to an accidental spill of fuel acute toxic effects on higher organisms in the water column may result from direct ingestion of the dissolved fraction and/or emulsified particles. On the other hand, benthic species may be affected later if and when a fraction of the fuel becomes entrapped in the sediment. After the passage of some longer time, a chronic effect may persist due to a food chain process, in which, for example, turbidicid worms may ingest hydrocarbon residues in the sediment and subsequently transmit these chemicals to higher trophic levels.

An issue that has received no mention to this point relates to advective transport of the fuel in a river or in tidal waters. A considerable literature exists on mathematical models of advective-dispersive transport and ecological/water quality models can be grafted onto these. The chemical transport and biological interaction portion of any such effort, however, are likely to inject much more uncertainty into projections than are physical mixing terms. In our opinion it is premature to consider the problems of including advective-dispersive terms in the model at this stage of the project since it is clearly a formidable undertaking to deal with the transport phenomena in a relatively quiescent situation. The present intention is to ignore advection (i.e., to consider a pond) which is in many instances a worst case situation.

In the above discussion various issues fundamental to the modeling exercise have been dealt with either implicitly or explicitly. The intention is to begin with a simulation model with twelve compartments. The model is a lumped parameter differential equation model based principally on mass balances between compartments. It is probably possible to deal realistically with volatilization and solubilization of the benzene, toluene, and xylene components but we foresee a good deal of interaction between modeling and experimentation as they relate to the emulsion problem. The first detailed effort will be devoted to the volatilization process.

The next phase of the modeling study pertaining to estimating the impact of spills of jet fuel, then, is to determine appropriate functional forms for the  $k_{ij}$  terms in the lumped parameter model. As indicated above, these are likely to be complex expressions and the resulting model will contain many uncertain parameters. Ranges for all parameters for the Monte Carlo simulations must also be gleaned from the literature to as great an extent as possible. If information on some particular aspect of the problem is unavailable (e.g., if some rate coefficient for volatilization of one of the aromatic components is not known), simple experiments will be designed and implemented to yield at least a crude estimate of associated parameters. After the model structure(s) to be investigated are identified and after parameter ranges have been specified, the generalized sensitivity procedure outline can be used to identify the uncertainties in the problem.



## APPENDIX A

### BIOLOGY OF Artemia

Brine shrimp Artemia salina L. is an anostracan phyllopod crustacean reaching lengths of 12 mm. It belongs to the same subclass Branchiopoda as the well-known cladoceran Daphnia but differs in having stalked eyes and lacking a carapace. Except for a few marine cladocerans, the brine shrimp is the only branchiopod inhabiting saline lakes or brine ponds throughout the world.

In optimum laboratory conditions Artemia reproduce every four to five days. A precopulation phase occurs when the male rides and clasps the female with its antennae between the uterus and the last pair of thoracopods. After copulation the female is capable of either ovoviviparous or oviparous reproduction. In some rare instances a female can simultaneously undergo both types of reproduction. Under ideal conditions, high oxygen levels, moderate temperatures and salinity, ovoviviparity occurs when after fertilization each of the eggs (20-150) is coated with a thin shell layer by the shell gland. The eggs hatch within five days in the female ovisac and then the nauplii are released into the environment. Oviparity involves eggs that have thick shell layers and is thought to be triggered by stressful conditions, such as low oxygen levels, starvation, and possibly high salinities (Bowen, 1962). Wheeler et al. (1979) found that the shell layer is composed of three distinct regions: 1) smooth outermost region consisting of outer membrane and cortical layer 1.2  $\mu\text{m}$  in thickness; 2) spongy middle region consisting of an alveolar layer 4.7  $\mu\text{m}$  in width; and 3) inner fibrous region (1.8  $\mu\text{m}$ ) composed of outer cuticular membrane, fibrous layer, and inner cuticular membrane. The hard brown external layer is lipoproteinaceous and contains hematin, a derivate of hemoglobin (Sorgeloos et al., 1977). Low oxygen level is proposed to increase hemoglobin production in the female and hematin, the degradation product of the pigment, is incorporated in the cyst shell. Mathias (1937) proposed that the middle alveolar layer contributes to the cyst buoyancy. Encysted embryos have their development stopped at the gastrula stage and will not ordinarily hatch unless the cysts have been desiccated. At this point, the dry cysts are deflated ball shaped about 0.18 mm in diameter. During two hours of rehydration the cyst rapidly absorbs water and becomes spherically shaped about 0.19 mm diameter. Clegg (1964) believes that oxidation of trehalose in the cysts contributes to the free glycerol production, creating an internal osmotic pressure for shell rupture and controlling possibly the rate of hydration.

Conte et al. (1977) describe the time period of the hatching process. The initial emergence after shell rupture is considered the early pre-nauplius stage ( $E_1$ ) and occurs after approximately 6-12 h after incubation. About 12 to 16 h postincubation the late prenauplius stage ( $E_2$ ) takes form as a teardrop shaped embryo surrounded by a hatching membrane and may or may not be attached to the cyst. Hatching is completed when the nauplius begins a series of intermittent beatings to break the hatching membrane and then swims freely away. The cross-shaped nauplius undergoes twelve molts and is sexually mature after two to three weeks. Adult brine shrimp can live up to six months under optimal laboratory conditions. They are

filter feeders and survive on unicellular algae and especially well on yeast.

Artemia is becoming increasingly important as a convenient food source for fish and crustacean culturists and aquarium hobbyists. Its cysts (dry eggs) can be transported to various parts of the world so mariculturists can hatch the eggs into larvae as food for their animals. In the United States the brine shrimp is harvested in South San Francisco Bay, California, and Great Salt Lake, Utah, and has been found in Los Angeles, San Luis Obispo, Mono Lake, Oregon, Nevada, New Mexico, and Texas. Much research has been done on the biology of Artemia. Sorgeloos et al. (1980) have compiled 2736 references on Artemia. The Artemia Reference Center in Belgium has been established as a research institute and information clearinghouse on the brine shrimp.

Brine shrimp are becoming important as a test species for toxicological studies (Tarpley, 1958; Leonhard and Lawrence, 1980). Michael et al. (1956) found that A. salina was superior to all other test organisms screened for bioassay testing; the advantages were that the animal was sensitive to wide ranges of compounds and that continuous culturing of animals was eliminated as stored cysts could be hatched within 24 h. Another advantage is that hatching can be synchronized by prehydration at low temperatures (3°C) for 4 to 10 h in distilled water (Conte et al., 1977).

The hatchability of cysts is also becoming a rapid test in assessing various toxicants. This method has been used in testing the effects of phthalate esters, organic solvents, and trace metals (Sugawara, 1974; Tazawa and Iwanami, 1974; Saliba and Krzyz, 1976). Kuwabara et al. (1980) have successfully tested 40 compounds with the cyst hatchability method. In addition, the cysts can be decapsulated by the method of Sorgeloos et al. (1977) to improve hatching numbers and to prevent possible introduction of bacterial growth and diseases in testing medium. The latter is of some concern since Wheeler et al. (1979) have observed microscopic bacterial flora on the outer shell layer.

APPENDIX B

ADDITIONAL BIBLIOGRAPHY PERTINENT TO MATHEMATICAL MODEL

# **Break Up of Oil on Rough Seas - Simplified Models and Step-by-Step Calculations**

Little (Arthur D) Inc Cambridge MA-Coast Guard, Washington, DC (208850)

Final rept.

AUTHOR: Aravamudan, K.; Raj, P. K.; Newman, E.; Tucker, W. A

G157184 Fld 139, 680 GRA18018

Oct 79 210p

Rept No: ADL-83066

Contract: DDT-CG-843466-A

Monitor: USCG-D-69-73

**Abstract:** This study tries to integrate the existing theoretical and experimental information regarding behavior of oil spills in ocean into a unified, sequential calculation procedure. Major consideration was given to developing the least sophisticated calculation procedures which maintain a reasonable description of the totality of the physical processes. The principal phenomena included in the prediction of spill behavior are: combined spreading and evaporation; the interaction of waves with the slick and formation of oil droplets; dispersion of droplets in the water column; and finally the formation and dispersion of slicklets due to turbulence in the ocean. The model has been exercised for four different types of oils: light crude, heavy crude, fuel oil no. 2 and fuel oil no. 6.

**Descriptors:** \*Oil spills, \*Water pollution, \*Oil pollution, \*Mathematical models, Dispersions, Drops, Ocean waves, Interactions, Fuel oil

Identifiers: NTISDDDA, NTISDOTOS

AD-A084 240-1 NTIS Prices: PC A10 MF A01

## **Summary of LNG Safety Research: Supplement - Task III**

Little (Arthur D.), Inc., Cambridge, Mass., Department of Transportation Washington, D.C. Office of Pipeline Safety, 1208 BSO

AUTHOR: Atallah, S.; Drake, E.; Reid, R

E016413 Fld 210, 13L, 97K, 94K GRA17202

Dec 74 208p

Rept No: ADL-C-76371-3

Contract: DDT-GS-40171

Monitor: 18

See also final rept., pg 241 042

**Abstract:** Results from major published research programs related to assessment and alleviation of potential LNG hazards are described. Topics covered include: vaporization and dispersion from LNG spills, boiling heat transfer rates for LNG on water, superheat explosions, tank stratification and rollover, thermal radiation from LNG pool fire, detonation conditions, pool spread and thermal radiation from ignited LNG

releases on water, deflagration of natural gas, air vapor clouds, and LNG fire control and suppression research. In addition to descriptive material, an extensive bibliography is included and future research needs are suggested

**Descriptors:** \*Liquefied natural gas, \*Safety, Vaporizing, Dispersion, Fire safety, Hazardous materials, Mathematical models, Fuel storage, Detonation, Water pollution, Air pollution, Flammability, Standards, Regulations, Environmental surveys, Explosions

Identifiers: Liquid wastes, Oil spills, NTISDOTOPS, NTISDOTOS

PB-273 378/OST NTIS Prices: PC A10/MF A01

## **Proceedings of the Workshop on Government Oil Spill Modeling Held at Wallops Island, Virginia on November 7-9, 1979**

National Oceanic and Atmospheric Administration, Washington, DC, Environmental Data and Information Service (031813056)

AUTHOR: Bishop, Joseph M.

G173134 Fld 139, 94L, 6C, 5F, 58D, 47, 98F, 57H, 86N GRA1

2019

Feb 80 220

Monitor: NOAA-80042401

**Abstract:** A Workshop on Government Oil Spill Modeling Activities was convened by the Environmental Data and Information Service (EDIS) of the National Oceanic and Atmospheric Administration (NOAA), November 7-9, 1979. The meeting was held at the National Aeronautics and Space Administration (NASA) training facility, at Wallops Island, VA. The Workshop brought together oil spill model users and modelers for the purpose of fostering joint communication and increasing understanding of mutual problems. The Workshop concentrated on defining user needs, presentations on ongoing modeling programs, and discussions of supporting research for these modeling efforts. Specific user recommendations include the development of an oil spill model user library, which identifies and describes available models. One user concern was the development of models for the long-term fate and effect of spilled oil.

**Descriptors:** \*Oil pollution, \*Water pollution, \*Meetings, \*Mathematical models, Research projects, Environmental impacts, Recommendations, Shores, Availability, Climate, Regions, Fisheries, Velocity

Identifiers: \*Oil spills, Water pollution effects (Animals), Oil spills, NTISCOMDA

PB-273 378 NTIS Prices: PC A10 MF A01

678545 ID NO. - EI761178545

**ANALYSIS OF AN EMULSIFIED OIL SLICK**

Collins, D. J.; Mackay, G. D. M.; Wong, K. T.  
Univ of Louisville, Ky

Offshore Technol Conf 8th Annu. Proc. Houston, Tex. May 3-6.  
1976 v 3 p 3 Pp OTC 2694 p889-839 CODEN: DSTCBA

The performance of mechanical nonporous booms as oil-spill containment devices has been studied previously, and the behavior of contained oil slicks has been observed and analyzed. These investigations are restricted to viscous oils that are Newtonian fluids. Some oils form water-in-oil emulsions, which are Bingham plastic and behave differently than Newtonian fluids. In this work, the stability of contained Bingham plastic oil slicks under the action of water current is investigated. The analysis is based on assuming a simple airfoil shape for the slick and calculating shear stress from boundary layer theory. The effect of dynamic forces on internal stresses is included in the study. The investigation shows Bingham plastic slicks to be more stable than viscous oil slicks. 14 refs.

DESCRIPTORS: (\*WATER POLLUTION, \*Oil Spills). MATHEMATICAL MODELS.

CARD ALERT: 453, 921

595213 ID NO. - EI751285213

**OIL SLICK FATE IN A REGION OF STRONG TIDAL CURRENTS.**

Drapeau, G.; Harrison, W.; Bien, W.; Leinonen, P.  
Univ du Que., Rimouski

Coastal Eng Conf, 14th Int. Proc. Copenhagen, Den, Jun 24-28 1974 v 3 p 2245-2259. Publ by ASCE, New York, NY, 1975

This study examines the drifting, spreading and aging of small slicks of crude oil in the middle St. Lawrence Estuary. This region was chosen because it is well documented with field measurements, hydraulic scale models, and mathematical models; and also because it is becoming a strategic area for the development of supertanker ports for 300,000 and possibly 500,000 ton tankers. The results indicate that it is impossible either to recover or to disperse small spills of oil in this region of strong tidal currents. Models also predict slick motion poorly. The alternative is to construct slick-drift roses that will indicate areas of expected beaching and assist in deployment of oil-spill clean-up technology. Data are presented in graphical form. 11 refs.

DESCRIPTORS: (\*WATER POLLUTION, \*Oil Spills). (TIDES, Models J, BEACHES.

CARD ALERT: 453, 407

321546 ID NO. - EI730421546

**PROPOSAL FOR THE APPLICATION OF MONOD'S MATHEMATICAL MODEL TO THE BIODEGRADATION OF MINERAL OIL IN NATURAL WATERS.**

Fasoli, U.; Nuemann, W.  
Politecnico Torino, Italy

Water Res v 7 n 3 Mar 1973 p 409-418 CODEN: WATRAG  
The study was designed to obtain a set of results that would serve as a means of investigating the possibility of employing

certain J. Monod equation in the interpretation mineral oil biodegradation; evaluation certain coefficients and determining their influence on the course of biodegradation; determining the part played by evaporation and re-oxygenation in the overall process of oil removal by comparison with the biological process, and the effect of the possible onset of anaerobic phenomena. 11 refs.

DESCRIPTORS: (\*WATER POLLUTION, \*Oil Spills). GAS OIL. MATHEMATICAL MODELS.

IDENTIFIERS: BIODEGRADATION

CARD ALERT: 453, 523, 921

**The Effects of Surface Phenomena on the Spreading of Oil on Water**

Massachusetts Inst. of Tech., Cambridge. Sea Grant Coll. Program, National Oceanic and Atmospheric Administration, Rockville, MD. Office of Sea Grant. (001450233)

AUTHOR: Fazal, Riyaz A.; Mitigrah, Jerome H.

G1211K1 F1d: 138, 68D, 86M GRA18014

Nov 79 73p

Rept No: MITSG-79-31

Grant: NOAA-04-7-158-44074

Monitor: NOAA-80021405

Abstract An oil spill in the ocean does not spread from its source evenly. Some components thin out into fine films, while others coagulate into thick patches. Most predictive models at present incorporate information on the bulk properties, but they do not account for this actual uneven fractionation. The oils are composites made up of many compounds which have different chemical and surface properties. This report describes studies of the spreading of four kinds of crude extracted from around the world, as well as the spreading of a mixture of pure hydrocarbons and of diesel fuel. It concludes that spreading effects are not wholly determined by bulk properties, but are also affected by the properties of the individual components in the oil.

DESCRIPTORS: \*Dispersing, \*Oil pollution, Mathematical models, Crude oil, Surface properties, Hydrocarbons, Diesel fuels, Mixtures, Decanes, Viscosity, Spreading, Interfacial tension, Thickness, Gas chromatography

Identifiers: \*Oil spills, Sea Grant program, NTISCOMNOA

P880-163629 NTIS Prices: PC A02/WF A01

1109721 ID NO. E1810109721  
DISTRIBUTION OF OIL POLLUTION RESIDUES

Furzer, Ian A.  
Univ of Sydney, Aust  
ASCE J Environ Eng Div V 106 n 6 Dec 1980 p 1171-1189  
CODEN: JEEGAV

ISSN 0090-3914

A new mathematical model is proposed for the simulation of the distribution of oil pollution residues along the length of a closed estuary. The model consists of a dual system of continuous stirred tank reactors with oil pollution residues initially suspended in the aqueous phase and being transferred to the sediments by settling. An effective numerical procedure was used to solve the system equations on a CYBER computer. Experimental measurements of oil pollution residues in the Parramatta and Duck Rivers, Australia can be modeled by this method. The parameters in the model were evaluated using a simplex optimization routine. The extensive experimental survey of these rivers indicate they are conservative. Contain 8,000 ton of oil pollution residues which will require several decades for complete oxygenation. The new modeling techniques proposed for the sediments are capable of handling single and multipoint sources of pollutants and will assist in water quality management of closed estuaries. 2 refs.

DESCRIPTORS: (WATER POLLUTION, Oil Spills), MATHEMATICAL MODELS, SEDIMENTATION, (RIVERS, Estuaries), IDENTIFIERS ESTUARIES.  
CARD ALERT 453, 931, 931, 407, 441

1053924 ID NO. E1800753924  
ASSESSMENT OF PREDICTABILITY OF LNG VAPOR DISPERSION FROM CATASTROPHIC SPILLS ONTO WATER.

Havens, Jerry A.  
Univ of Ark, Fayetteville  
J Hazard Mater V 3 n 3 Jan 1980 p 257-278 CODEN JHMAD3  
ISSN 0304-3894

Predictions of the downwind travel of LNG vapor air mixtures based on mathematical modeling by several groups have shown wide variations. A review and assessment of these predictions is summarized in this paper. Its objectives were to provide a detailed description of the models, and using them to estimate maximum downwind travel for a standard scenario. To enable valid comparisons to be made, to identify the reasons for differences found, to define the present state of the art, and to recommend further state-of-the-art improvements. Seven groups and the predictions which they give, are described and compared. 12 refs.

DESCRIPTORS: NATURAL GAS, LIQUEFIED, (VAPORS, Diffusion), IDENTIFIERS WATER POLLUTION, & OIL SPILLS  
CARD ALERT 512, 522

## Methodology, Evaluation and Appendices

Mathematical Sciences Northwest Inc Bellevue Wash-Coast Guard.  
Washington, D.C. Office of Research and Development. 1404058

Final rept.

AUTHOR: Isakson, J. S.; Storrie, J. M.; Vagners, J.; Erickson, G. A.; Kruger, J. F.  
F5593d4 FID 13B, FR. #90-. 57H-. 47 GRAI7601

Jun 75 633p

Contract D01-CG-42913-A

Monitor: USCG-D-155-75-Vol-1

See also volume 2. AD-A017 501

Abstract: A ranking of potential environmental impact for spills of crude oil, diesel-2, bunker C, and gasoline in amounts ranging from 100 to 50,000 barrels was made for specific sites at Iktuat Bay, Valdez Harbor, Valdez Narrows, Drift River Channel, Port Graham, Kamishak Bay, Unimak Pass, Port Moller, Kvichak Bay, St. Matthew Island, Offshore Prudhoe, Onshore Prudhoe, Nome, Cape Blossom Channel, Colville River, River, In River, and Denali Fault. Spills were assumed to disperse from inertial, viscous, surface tension, wind and current forces. Most probable wind and current conditions were utilized. Sites were characterized in terms of eight species hat rats. Most probable cases were evaluated. A rating system was devised to characterize the impact based upon estimated species abundance, importance of species, and the impact of oil on such species over the short and long term. Impacts were estimated with the use of three-dimensional matrices. The same five locations dominated the impact ratings where containment/cleanup were assumed to take place.

Descriptors: Oil spills, Ecology, Alaska, Oil pollution, Sires, Invertebrates, Plankton, Fresh water, Mixing, Fishes, Climatic, Vegetation, Petroleum products, Removal, Aquatic plants, Ocean currents, Wind, Abundance, Ratings, Computerized simulation, Mathematical models, Habitability, Rivers, Harbors, Marine terminals, Gasoline

Identifiers: Environmental impacts, Evaluation, Water pollution effects (Animals), Crude oil, Water pollution effects (Plants), Oil pollution removal, NTISDDCG NTISDDCG

AD-A017 400, 851 NTIS Prices PC F09/MF 401

310781 ID NO. E1730210781

## INSTABILITY AT AN INTERFACE BETWEEN OIL AND FLOWING WATER.

James, W. T.

Shell Pure Line Corp, Houston, Tex

ASME Pap n 72-WA/Oct-7 for Meet Nov 26-30 1972, 5 p

A linear, inviscid stability analysis is applied to the case of a relatively thin layer of stationary oil over a semi-infinite layer of water flowing at a constant velocity. Predictions of the velocity, at which the interface becomes

Comparison of Ecological Impacts of Postulated Oil Spills at Selected Alaskan Locations, Volume I. Introduction. Summary.

unstable and effect of the oil depth on this velocity agree qualitatively with observations. Results can be applied to the problem of containing oil with mechanical booms. The model predicts initiation of oil loss by droplet separation from beneath the slick at low current velocities. For example, 0.39 to 0.53 fps for No. 2 diesel fuel depending on slick thickness. 13 refs.

DESCRIPTORS: (\*WATER POLLUTION, \*Oil Spills), OIL BOOMS, MATHEMATICAL MODELS.

IDENTIFIERS: OIL-WATER INTERFACE

CARD ALERT: 453

# **Workshop on the Physical Behavior of Oil in the Marine Environment, Held at Princeton University, Princeton, New Jersey, on May 8-9, 1979**

Princeton Univ., NJ. Dept. of Civil Engineering. \*National Weather Service, Silver Spring, MD. Techniques Development Lab. (009938013)

AUTHOR: Kerr, Christopher L.; Barrientos, Celso S. G3244L4 Fld: 13B, 48, 8C, 68D\*, 55C, 47B, 86T GRA18105 1979 386p\*

Monitor: NOAA-80100815

**Abstract** A workshop on the Physical Behavior of Oil in the Marine Environment, was held at Princeton University, New Jersey on May 8-9, 1979. Workshop presentations were grouped into the following sections: experimental study, theoretical research, operational forecasts, risk assessment, physical behavior in cold regions and oceanography and meteorology.

DESCRIPTORS: \*Forecasting, \*Oil pollution, \*Meetings. Mathematical models, Emulsions, Crude oil, Risk, Ocean currents, Dispersion, Arctic regions, Trajectories, Air water interactions, Dynamics, Sea ice, Flow charting, Ocean waves, Viscosity, Density(Mass/volume), Weather observations

Identifiers: \*Oil spills, Oil slicks, NTISCOMNDA

P881-123796 NTIS Prices: PC A17/MF A01

# **Modeling Methods for Predicting Oil Spill Movement**

Oceanographic Inst. of Washington, Seattle \*Oceanographic Commission of Washington, Seattle. (405 972)

AUTHOR: Rath, Robert J.; Francis, Bradley H.

E1025H4 Fld: 13B, 68D\*, 47 GRA17811 Mar 77 106p\* Rept No: OIW/OCW-7502 Monitor: 18

**Abstract** The study consists of a literature search and an evaluation of 7 representative papers on modeling the surface movement of spilled oil in the marine environment. The modeling approaches vary considerably in complexity and are based on experiments, observations, and theoretical considerations, or combinations thereof. In addition to the evaluation of the individual models, the study contains an overview of the state-of-the-art of predicting oil spill movement by utilizing oil spreading and drift models. The overview includes a discussion of controversial parameters, methods of model verification, and studies which are being conducted to extend the methodology to three dimensions by considering the distribution of spilled oil in the atmosphere, the water column, and bottom sediments.

DESCRIPTORS: \*Mathematical models, Water pollution, Transport properties, Forecasting, Drift, Field tests, Abstracts, Trajectories

Identifiers: \*Oil spills, \*Oil spreading process, NTISOCW

PS-277 669/85T NTIS Prices: PC A06/MF A01

# **A Critical Technical Review of Six Additional Hazard Assessment Models**

Enviro Control Inc Rockville Md (408195)

Final rept.  
AUTHOR: Rausch, A. H.; Kumar, R. M.; Lynch, Cornelius J. D3582D1 Fld: 13B, 68D GRA17724 Mar 77 205p Contract: D07-CG-33377-A Monitor: USCG-D-54-77 See also Rept. no. USCG-D-136-75, AD-A015 245.

**Abstract:** This report presents a critical, technical review of six simulation models currently being used in connection with the following U.S. Coast Guard research programs: the Vulnerability Model, a simulation system for assessing damage resulting from spills of hazardous chemicals; and CHRIS, the Chemical Hazards Response Information System. (Author)

DESCRIPTORS: \*Water pollution, \*Coast Guard research, \*Computerized simulation, \*Mathematical models, Hazardous materials transportation, Chemical contamination, Oil spills, Damage assessment, Environmental protection, Research management, Documents, Liquids, Vapors, Dispersions, High viscosity, Management information systems, Cryogenics

Identifiers: Chemical hazards response information system, \*Reviews, NTISD0D>A

AD-A044 198 /OS, NTIS Prices PC A10/MF A01

# Confined Boiling Rates of Liquefied Petroleum Gas on Water

Massachusetts Inst. of Tech., Cambridge, Dept. of Chemical Engineering, Department of Energy, 13710000  
 AUTHOR: Reid, R. C.; Smith, K. A.  
 E270414 Fld. 70, 680, 99F GRA17825  
 May 78 51p  
 Contract: EE-77-S-02-4548  
 Monitor: 18

**Abstract:** Results of a program to measure the rate of boiling of liquefied petroleum gas (LPG) on water surface and to develop an analytical model to describe the phenomena involved are reported. Primary emphasis was placed on liquid propane or LPG mixtures containing small quantities of ethane or butane or both. A few exploratory tests were, however, made with pure liquid ethane, ethylene, and n-butane. The investigation was conducted to provide quantitative data and analytical models to delineate the rate of vaporization, the spread rate and the degree of fractionation, should an LPG tanker suffer an accident leading to a major spill on water. For propane or LPG spills on water, immediately following the contact, violent boiling commenced. Ice quickly formed; in most cases, ice was even thrown onto the sidealls of the vessel. In some instances sprays of water/ice and propane were ejected from the calorimeter. Within a few seconds, however, the interaction quieted and the surface was covered by a rough ice sheet. The LPG boiled on the surface of this ice, but large gas bubbles occasionally appeared under the ice shield and were trapped. The boiling rate decreased with time with a concomitant increase in the thickness of the ice shield. In the first second or two, very high boiling heat fluxes were experienced. The mass of LPG lost was approximately half that spilled originally. It is estimated that only 5 to 15% could have been ejected as liquid if the water loss is used as a reference. However, since the water surface is very agitated during this period, it is not possible to obtain reliable quantitative values of the boiling flux. Also, as noted, the mass lost in the very early time period was approximately proportional to the original mass of LPG used. It may be inferred that larger spills lead to more mixing and boiling before the ice shield prevents a direct contact between the LPG and the water. (ERA citation 03 046762)

**Descriptors:** Gas spills. Liquefied petroleum gases. Tanker ships. Boiling. Butane. Ethane. Ethylene. Evaporation. Fractionation. Propane. Water

**Identifiers:** ERDA/O30800. Mathematical models. Oil spills. Oil pollution. NTISDE

HCP/P4548-01 NTIS Prices PC A04 MF A01

603109 ID NO. E1760103109

## EFFECTS OF TEMPERATURE ON OIL REFINERY WASTE TOXICITY.

Reynolds, James H.; Middlebrooks, E. J.; Porcella, D. B.; Greeney, W. J.  
 Utah State Univ., Logan  
 J Water Pollut Control Fed v 47 n 11 Nov 1975 p 2674-2693  
 COOFN: JMFPA5

A continuous flow kinetic model was developed to describe and predict the effects of temperature on the toxicity of a specific oil refinery waste to the green alga, *Selenastrum capricornutum*. The model is based on Michaelis-Menten enzyme inhibition kinetics, with ammonium nitrogen-limited, continuous flow algal cultures between 20 degrees and 28 degrees C. Phenol is the controlling inhibitor. The model was applied to continuous flow algal cultures exposed to an actual oil refinery waste. 70 refs  
**DESCRIPTORS:** (INDUSTRIAL WASTES. Treatment). (PETROLEUM REFINERIES. Wastes). (WATER POLLUTION. Oil Spills). (WATER ANALYSIS. Toxicity). MATHEMATICAL MODELS. IDENTIFIERS: ALGAL CULTURES  
 CARD ALERT 445, 452, 453, 513, 801, 921

770479 ID NO. E1770970479

## REVIEW AND EVALUATION OF BASIC TECHNIQUES FOR PREDICTING THE BEHAVIOR OF SURFACE OIL SLICKS

Stolzenbach, Keith D.; Madsen, Ole S.; Adams, E. Eric; Pollock, Andrew M.; Cooper, Curtis K.  
 MIT Water Resour & Hydrodyn. Ralph M. Parsons Lab  
 MIT Dep Civ Eng Ralph M. Parsons Lab Water Resour Hydrodyn Rep n 222 Feb 1977 322 p CODEN: MHVTAK

The purpose of the study described is to conduct a state-of-the-art review of the basic techniques and knowledge associated with surface oil slick behavior. This report presents the results of this review. Because of its significance in determining the fate of oil slicks, the representation and determination of wind fields is discussed first. The report presents a treatment of the advection of oil slicks that is brought about by the combined action of currents and wind waves and deals with the various physical phenomena that transform an oil slick as it is being advected. A review and evaluation of existing models for oil slick behavior is also presented. A comprehensive bibliography is included along with an appendix containing a detailed treatment of wind driven currents. 262 refs.  
**DESCRIPTORS:** (WATER POLLUTION. Oil Spills). WIND EFFECTS. WATER WAVES. MATHEMATICAL MODELS. IDENTIFIERS: WIND DRIVEN CURRENTS  
 CARD ALERT 443, 453, 471, 671, 921



127499 \*78-002471

**A REVIEW AND EVALUATION OF BASIC TECHNIQUES FOR PREDICTING THE BEHAVIOR OF SURFACE OIL SLICKS.**

STOLZENBACH, KEITH D. ; MADSEN OLE S ; ADAMS E. ERIC ; POLLACK ANDREW M. ; COOPER CORTIS K. MIT.

MIT SEA GRANT PROGRAM REPORT MITSG 77-8. MAR 77 (322)  
SPECIAL REPORT: THE BASIC ANALYTICAL TECHNIQUES AVAILABLE FOR PREDICTING THE FATE OF OIL SPILLS ARE REVIEWED IN DETAIL. MAJOR FACTORS CONSIDERED ARE: THE LOCATION, SIZE, AND PHYSICAL AND CHEMICAL PROPERTIES OF THE OIL SPILLED; TRANSPORT OF THE OIL BY WIND AND CURRENTS; AND THE PHYSICAL, BIOLOGICAL AND CHEMICAL TRANSFORMATIONS THAT THE OIL UNDERGOES AS IT IS BEING TRANSPORTED. THE STATE OF THE ART OF MODELING TECHNIQUES AND KNOWLEDGE ASSOCIATED WITH OIL SLICK BEHAVIOR IS SURVEYED. EXISTING MATHEMATICAL MODELS ARE EXAMINED. AND RESEARCH NEEDS FOR BETTER MODELING EFFORTS ARE DISCUSSED. (NUMEROUS DIAGRAMS, GRAPHS, REFERENCES, TABLES)

DESCRIPTORS: \*HYDROCARBONS-WATER ; \*OIL SPILLS ; \*WINDS ; \*BIODEGRADATION-MICROORGANISM ; \*EVAPORATION ; \*EMULSIFICATION ; \*MATHEMATIC MODELS-WATER ; NEW YORK BIGHT ; DELAWARE ; SAN FRANCISCO

REVIEW CLASSIFICATION 12

**Mathematical Description of Adsorption and Transport of Reactive Solutes in Soil: A Review of Selected Literature**

Oak Ridge National Lab., TN. Department of Energy. 14832000)  
AUTHOR: Travis, C. C.  
F099502 Fld. RM, 68 GRA17912  
Oct 78 83p  
Contract W-7405-ENG-26  
Monitor 18

Abstract: This report reviews selected literature related to the mathematical description of the transport of reactive solutes through soil. The primary areas of the literature reviewed are (1) mathematical models in current use for description of the adsorption-desorption interaction between the soil solution and the soil matrix and (2) analytic solutions of the differential equations describing the convective-dispersive transport of reactive solutes through soil. (ERA Citation 04 0170791)

Descriptors: \*Adsorption, \*Desorption, \*Soils, Convection, Differential equations, Dispersion relations, Solutions, Analytical solution, Environmental transport, Fertilizers, Ground water, Liquid wastes, Mathematical models, Oil spills, Pesticides, Radioactive wastes, Radionuclide migration, Reviews, Sorptive properties, Water

Identifiers: ERDA/510200, ERDA/510301, ERDA/510500, Environmental surveys, Path of pollutants, NITSOE

ORNL-5403 NIS prices PC A05 MF A01

983764 ID NO. - E1791083764

**OIL SPILLS IN A RIVER: A ONE-DIMENSIONAL MODEL.**

Voznesensky, G. Th  
Inst of Appl Geophys., Moscow, USSR  
Hydrol Sci Bull Sci Hydrol v 24 n 2 Jun 1979 p 213-223  
CODEN: HYSEAY

A linear one-dimensional model is proposed to describe the river pollution resulting from oil spills. Two interacting migratory forms, water surface pollution and water body pollution, are considered. In the coordinate system moving with the mean flow velocity, the equations for the pollutant distributions are reduced to the type of a heat conduction equation with distributed unsteady sources. The solutions for oil concentrations, obtained in the form of the 'heat potential', are considered in two particular cases: an oil spill in an open channel and river pollution from drifting ice due to the melting of polluted ice. 7 refs.

DESCRIPTORS: (-WATER POLLUTION, \*Oil Spills), MATHEMATICAL MODELS, (FLOW OF WATER, Open Channels), IDENTIFIERS: RIVER POLLUTION  
CARD ALERT: 453, 407, 444, 521, 631

**Modeling of U. Evaporation in an Aqueous Environment. Research on the Effects of Crude Oil Transfer and Upstream Refineries on Delaware Bay**

Delaware Univ., Newark. Coll. of Marine Studies. \*Delaware Univ., Newark. Dept. of Civil Engineering. \*National Science Foundation, Washington, D.C. Research Applied to National Need. (A07 178)

AUTHOR: Wang, Hsiang; Yang, Wei C.; Huang, C. P.

D236244 Fld. 13B, 68D GRA17713

Dec 76 42p

Rept No. CMS-RANN-5-76

Monitor NSF-GI-41896

See also PB-262 727.

Prepared in cooperation with Delaware Univ., Newark. Dept. of Civil Engineering. Rept. no. Ocean Engineering-7.

Abstract: The goal of the overall research program is to provide information on the input of oil transfer operations in Delaware Bay for decision makers. The work reported here consists of the computer modeling of oil weathering in the early stage of a spill. A numerical model is developed to predict changes in oil characteristics, specific gravity, residues by weight and residues by volume of oil on water through evaporation known to dominate the early stage of weathering. The major driving forces for evaporation being considered include air temperature, wind speed, and size of slick. Laboratory experiments were conducted using No. 2 fuel oil, Nigerian Crude, and Venezuelan Crude to calibrate the numerical model and to gain physical insight into the oil evaporation process. Based on the laboratory results, empirical relationships are proposed for determining the

diffusion coefficient described in the first-order decay formula. Both numerical and laboratory results seemed to indicate that the effect of temperature on oil weathering is significant in the very early stage of weathering, but diminishes at a later time. The effect of wind speed is more uniform throughout the time duration being tested

Descriptors: \*Crude oil. \*Ocean environments. \*Oil pollution. Delaware Bay. Mathematical models. Computerized simulation. Weathering. Evaporation. Residues. Atmospheric temperature. Refineries. Wind velocity. Size determination. Fuel oil. Diffusion. Test methods. Decision making

Identifiers: \*Oil spills. Oil slicks. NTISNFR

PB 264 968/95 NTIS Prices PC A03/MF A01

814733 ID NO. - EI780214733

#### MODELING OF OIL EVAPORATION IN AQUEOUS ENVIRONMENT.

Yang, Wei C.; Wang, Hsiang

Technische Univ. Braunschweig, Ger

Water Res. V 11 N 10 1977 p 879-887 CONF. MATRAG

A numerical model is developed to predict the changes of oil characteristics, specific gravity, percentage weight remain and percentage volume remain of oil on water through evaporation known to dominate the early stage of weathering. This model is based on the assumption that oil, though complex, can be considered as a chemical compound of individual fractions of distinguishable chemical and physical characteristics. The major driving forces for evaporation being considered include air temperature, wind speed and size of slick. Laboratory experiments were conducted using No. 2 fuel oil, Nigerian Crude and Venezuelan Crude to calibrate the numerical model and to gain physical insight of the oil evaporation process. Based on the laboratory results, empirical relationships are proposed for determining the diffusion coefficient described in the first-order decay formula. 10 refs

DESCRIPTORS: (\*WATER POLLUTION. \*Oil Spills). EVAPORATION. MATHEMATICAL MODELS.

IDENTIFIERS: AQUEOUS ENVIRONMENT. DIFFUSION

CARD ALERT 453, 802, 921

#### A Numerical Model of Droplet Entrainment from a Contained Oil Slick

Mount Auburn Research Associates Inc. Newton Upper Falls Mass-Coast Guard. Washington, D.C. Office of Research and Development. (390072)

Final report

AUTHOR: Zaioch, Robert G.

C441222 Fld 200, 68D, 468 GRA17510

Sep 74 83p

Contract: D01-CG-41822-A

Monitor USCG-D 55-75

Abstract: A theoretical analysis of oil droplet entrainment from a contained oil slick moving relative to water has been performed as a function of relative oil-water velocity. A numerical method incorporating discrete vortices is used to calculate smooth stable headwave profiles at low velocities and unstable profiles at high velocities. An oil droplet formation criterion is formulated and applied to the numerically modeled headwave region. The computed critical velocity corresponding to the onset of significant droplet entrainment is in close agreement with recent laboratory measurements. The computed entrainment rates are in approximate agreement with experiment, but do not exhibit systematic variation with water current. Oil drop trajectories are calculated using realistic starting conditions, but the present work does not extend beyond the first intersection of the drop with the slick. Recommendations are given for extending the work to include post-formation droplet dynamics, wave effects, turbulence effects, and barrier design changes.

Descriptors: \*Drops. \*Entrainment. Water flow. Rates. Oil spills. Mathematical models. Vortices. Computer programs. FORTRAN

Identifiers: \*Oil slicks. FORTRAN 4 programming language. Oil pollution containment. NTISDDCG. NTISD01

AD A-006 600/157 NTIS Prices PC A05/MF A01

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